Spatial variation in growth and survival of juveniles of the American lobster, *Homarus americanus*, along the Maine coast

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by

Brian F. Beal, Director of Research
Downeast Institute for Applied Marine Research & Education

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Summary

A two-year field investigation was conducted at six sites along a 340 km stretch of the Maine coast to examine spatial variability in growth and survival of juvenile American lobsters, *Homarus americanus*. To enable unambiguous size-at-age estimates, lobsters were cultured from broodstock from a single origin during the summer of 2006. At each site, an individual stage IV-V postlarvae (mean carapace length [CL] ± 95% CI = 4.1 ± 0.05 mm) was placed into one of two sizes of plastic containers (3.3 L with bottom surface area of 308 cm^2; 4.2 L with bottom surface area of 230 cm^2) that entrapped the animal, but allowed seawater to flow through the space. A microhabitat, consisting of large pieces of crushed shells of adults of the soft-shell clam, *Mya arenaria*, was added to the bottom of one-half of both sizes of containers to a depth of 3 cm. Containers were placed in groups of twelve within wire cages the size of standard commercial lobster traps. Cages were connected to each other in groups (blocks) of four with 2-3 m spacing between adjacent cages, and then four blocks were deployed on both a hard and soft bottom at each study site (number of lobsters = 6 sites x 4 blocks x 2 habitats x 2 container types x 2 microhabitats x 12 containers/cage/block = 2,304 lobsters). One cage in each block was assigned to each of the four treatments (a factorial combination of bucket size [a = 2] and microhabitat [b = 2]). This was a traditional randomized complete block design. Survival estimates were recorded in October 2006 at each site, and in June 2007 at five of the six sites. At the end of the study (August 2008), carapace length and wet mass of each live lobster was recorded.

Overall mean survival was 62.5% in October 2006 and 46.5 ± 5.5% in June 2007. Of the 192 cages deployed in June 2006, 114 were recovered in June 2007, and only 76 (39.6%) were recovered in August 2008. Storms, fishing-related gear entanglements, and other events were responsible for the loss of equipment. Only 338 live lobsters (14.7%) were recovered, which resulted in a final mean survival estimate of 37.1%. Final mean number of lobsters recovered was site-specific, with the highest recovery rate occurring in Cutler, where all gear was deployed in the harbor (51.2 ± 8.2%, n = 27), and lowest at York (12.0 ± 4.7%, n = 9). Mean number recovered in 2008 was approximately 35% higher on hard bottoms than soft bottoms ($P > 0.05$), which was opposite from what was observed in 2007, when lobsters recovered from soft bottoms was 52% greater than from hard bottoms (54.6 ± 6.9%, n = 64 vs. 36.1 ± 8.3%, n = 50). Neither

container size nor microhabitat had a significant effect on number of lobsters recovered in June 2007 or August 2008.

Final mean CL was relatively small, and varied across sites, but not as expected given that eastern Maine waters are cooler, on average, than the waters along the southwestern coast. Greatest mean CL was observed at Beals and Stonington where lobsters attained a similar final length (mean CL = 14.3 ± 0.77 mm, n = 15 cages) that was nearly 35% larger than lobsters from the remaining four sites (mean CL = 10.7 ± 0.34 mm, n = 61 cages). Effects of container size and microhabitat on final mean CL were statistically significant at only two sites, Cutler and Tenants Harbor. At each, lobster juveniles were larger in containers with the greatest surface area with shell microhabitat (by 8.7% in Cutler, and 25.8% in Tenants Harbor). No significant differences in mean CL were observed at either site for animals in containers without microhabitat.

Additional field tests demonstrated that lobster growth increases with size of container, so it is difficult to know whether the growth estimates observed here are representative of the population of wild juveniles of comparable sizes. The range of final mean CLs (5.9 – 19.1 mm) is quite variable, but the maximum value is within the range of estimates for two year-old juveniles reported elsewhere along the Maine coast. Future field investigations to determine growth-at-age should examine effects of: 1) container size/shape over a wider range of sizes; 2) water depth within a given location; 3) habitat stability within a given location; and 4) initial lobster size on growth trajectory.

Introduction

The American lobster, *Homarus americanus* Milne Edwards, is the most important commercially harvested marine species along the coast of Maine. In 2007, for example, nearly 63.1 million pounds were landed in Maine worth $280 million (http://www.maine.gov/dmr/commercialfishing/documents/2007LandingsBySpecies.Table.pdf). In addition, the State of Maine sells approximately 7,000 lobster licenses, making this fishery not only the most valuable, but the most popular form of commercial endeavor along the coast.

Like all crustaceans, lobsters grow by shedding their old shells, a process known as molting, or ecdysis. During the molt, lobsters leave no record of age or previous size because they lack permanent hard structures, and this makes understanding growth and estimating age-structure in crustacean populations difficult. Tagging studies (Cowan and Ellis 2000) have revealed a high degree of variability in lobster size after a period of time. For example, Cowan and Ellis (2000) reported a case study of six lobsters caught four times in the vicinity of Lowell's Cove, Orr's Island, in Casco Bay. One lobster, tagged initially on 23 July 1994 and measuring 18 mm carapace length (CL), grew approximately 5 mm over the next 11 months. From 14 June 1995 to 9 September 1995, that individual molted twice reaching a CL of 31 mm, an increase of nearly 35%. The next time the same lobster was caught (270 days later on 6 June 1996), it still had a CL of 35 mm. Other lobsters in that study of similar initial sizes molted 2-3 times per year, mostly during summer months. One problem with studies of this nature, however, is that tagging is inefficient (tags are lost and so, too, is the information for an individual's growth), and the age of the tagged animal is unknown – all one knows is the change in CL during the period between release and recapture. Unless the exact age of the animal is known at the time of tag-ging, this method is an imprecise one for estimating lobster age. In addition, behavior and/or growth may be influenced by the presence of the tag or the tagging process (Comeau and Savoie 2001).

Carapace size in crustaceans is influenced by a variety of abiotic and biotic factors such as salinity, temperature, molting frequency, food quality, and life stage of the individual (Sheehy et al. 1996; Ju et al. 2001). Without knowing these characteristics, it is difficult to estimate lobster

age from a distribution of sizes, although there are several statistical approaches that involve modal analysis of length-frequency data (e.g., Castro 1995; Robinson and Tully 2000).

An alternative approach to aging lobsters and other crustaceans involves using biochemical and histological techniques to quantify the autofluorescent age pigment, lipofuscin (Sheehy et al. 1996; Wahle et al. 1996; Ju et al. 2001) in individual animals. Accumulation of the pigment occurs over the lifespan of an individual and seems to be a universal correlate of animal senescence. Lipofuscin (LF) is a conglomerate of lipids, metals, organic molecules, and biomolecules that fluoresces at 360-470 nm. LF granules have been found in every eukaryote ever examined, and always accumulate within cells as the organism ages (Gaugler 1997). There are two ways to calibrate lipofuscin concentration against age: 1) analysis of a group of individuals of known age, covering as much of a species' lifespan as possible; or, 2) modal separation of a lipofuscin-frequency distribution (Sheehy et al. 1998).

Although the technique has shown promise in both European and American lobsters, formation and accumulation of lipofuscin may be affected by spatial and temporal environmental variability (Sheehy et al. 1996; Bluhm et al. 2001). In addition, lipofuscin accumulation in eyestalks, brains, and other lobster tissues may vary with temperature. For example, O'Donovan and Tully (1996) found significant differences in lipofuscin accumulation in European lobsters, *Homarus gammarus*, kept at 8^{o}C vs. 13^{o}C. Food availability, and therefore caloric intake, also affects the lipofuscin formation process (Chapelle et al. 1994). Besides environmental variability, genetically determined factors inherent to a species and/or individual, such as the activity level or feeding type, can affect lipofuscin accumulation (Bluhm et al. 2001). These studies suggest that without knowing the initial age of individuals, what temperature regime(s) individuals were exposed to, and/or the type and amount of food ingested, that the lipofuscin accumulation technique may have too many assumptions to make it practical or appropriate in a fisheries management setting.

One aspect of aging lobsters is clear. If one begins with known-age individuals, there is no ambiguity about age/size relationship after some period of time (t). Here, I report results from a two-year study conducted over a 340 km stretch of the Maine coast at six study sites from Cutler

(easternmost) to York (westernmost). At each site, I examined interactive effects of habitat (soft- vs. hard-bottom), microhabitat (shell vs. no shell), and the size of the container used to house lobsters. I used cultured juveniles (Beal and Chapman 2001) from parents from a single location to reduce potential variability due to genetic differences among broodstock. Juveniles (stages IV-V) were housed individually in plastic containers designed to allow a constant exchange of seawater. Animals survived and grew by feeding on detrital material and organisms that settled into and onto the hard surfaces of the containers (see Beal et al. 2002). Site- and treatment-specific size-frequency distributions can be used to compare with distributions of wild-caught juveniles (e.g., Wahle and Incze 1997; Cowan et al. 2004), providing a management tool to help interpret modal analyses used to estimate age distribution in wild juvenile lobster populations.

Methods and Materials

Origin of animals

This study investigated spatial variation in survival and growth of early benthic phase American lobsters, *Homarus americanus*. To remove ambiguity regarding initial age/size of experimental animals, cultured individuals were used in all trials. Stage I larvae were obtained from broodstock collected from Beals, Maine (Western Bay) beginning in mid-June 2006. Larvae were reared communally in 400-liter conical tanks at the Downeast Institute for Applied Marine Research and Education (DEI; Beals, Maine; 44°28.83'N; 67°35.90'W) according to Beal and Chapman (2001). Until used in field experiments, stage IV-V juveniles were held individually in plastic, flow-through compartments (ITML® horticultural tray; product code: INP32WPD, http://www.itml.com/prodDetail.php? pd=984) that floated in a 35,000 liter tank at DEI receiving ambient seawater (Table 1; Fig. 1). Animals were fed every other day with live brine shrimp, *Artemia salina*, that had been enriched with cultured microalgae (T. *Isochrysis galbana*; *Chaetoceros mulleri*).

Flow-through containers

Early benthic phase, cultured lobsters survive and grow *in situ* when placed individually into containers that permit flow (seawater exchange), but entrap the animal (Beal et al. 2001). Lobsters feed by cropping/grazing organisms and macroalgae that recruit and settle on the inside of the container or that drift into the container from the water column. Previous work suggested that amount of flow was important for long-term survival. For example, when lobsters were added to 15 cm x 2.5 cm Petri dishes in which approximately 32% of the top and bottom of the dish was replaced with a piece of nylon window screening, survival in submerged cages over an 11-month period near Beals, Maine was 91.7 ± 5.3% (n = 18) compared to 74.9 ± 8.6% (n = 12) for animals housed in similar dishes with 25 small holes (ca. 3 mm) drilled in the top and bottom cover of the dish (Beal 2006).

In the present study, lobsters were individually housed in two different size/shape round, plastic (food-grade, polypropylene, Bisphenol A-free) containers (see http://www.ipl-plastics.com).

The first was a “Squat bucket” (model 3012; white, 3.3 L capacity; 0.097 wall thickness; 19.8 cm diameter x 15 cm tall). The other was a “Tall bucket” (model 3712; white, 4.2 L capacity; 0.097 wall thickness; 17.1 cm diameter x 19.8 cm tall). To ensure ample flow, a hole (11.4 cm diameter) was cut from the bottom of each container, and replaced with a piece of nylon window screening (aperture = 1.8 mm) that was affixed to the remaining bottom lip using hot glue (general purpose, multi-temp glue stick, see http://www.glu-stix.com; Fig. 2a). In addition, each container had a polypropylene lid that when pushed down over the upper lip of each container formed a tight seal. A hole of similar diameter was cut in each lid, but instead of hot-gluing a piece of window screening to fill the hole, a piece (23 cm x 23 cm) of window screening was used to cover the top of the open container. The lid was then pushed over the rim to secure the screening. This arrangement (Fig. 2b) allowed seawater to flow into and out of each container.

Submerged cages

Cages (PVC-coated, galvanized 14-gauge wire, 81.2 cm x 45.7 cm x 30.5 cm with 2.54 cm apertures) similar to a standard, commercial, in-shore lobster trap, and with cement ballast, were used to house twelve flow-through containers. Each cage was fitted with a door (81.2 cm x 40 cm) that allowed access to the cage interior. A total of eight Squat buckets could be arrayed on the bottom of a single cage. A piece of coated wire (40 cm x 30 cm) was placed on top of each group of four buckets, and was secured in place using nylon cable ties. The wire pieces provided a solid surface upon which to place the four remaining buckets (Fig. 3a). All twelve of the Tall buckets could be arrayed on the bottom of a cage (Fig. 3b).

Study sites and lobster handling prior to initiating field trials

Six study sites were selected along the Maine coast to maximize geographic data regarding growth and survival of juvenile lobsters (Table 2). On each initiation date, approximately 500 stage IV-V lobsters were removed from their individual flow-through compartments at the Downeast Institute, and placed onto wet paper toweling within a stainless steel sieve (ca. 100/sieve). Sieves were then stored in plastic coolers containing several blocks of Rubbermaid

Blue Ice®. To ensure that lobsters were handled similarly and that they remained out of seawater for the same length of time regardless of distance traveled to a site, animals were held in coolers for five hours before handling them at a particular study site. One commercial fisherman from each of the six areas participated in this research. Each fisherman used his fishing vessel to hold/transport gear and lobsters, and assisted in carrying out the experimental design (see below). To minimize handling mortality, at each study site groups of 25-30 lobsters at a time were carefully removed from one of the sieves on board the fishing vessel by gently dipping an edge into a shallow enamel pan containing ambient seawater. Vigorous (actively swimming or crawling) individuals were "captured" using an 80 ml beaker. A single lobster with both claws was then placed (gently poured) into the bottom of a Tall or Squat bucket before the cage was placed overboard.

Experimental design

The experimental design focused on answering three questions at each site: 1) Does lobster survival and growth vary between soft- and hard-bottom habitats? 2) What effect does container size have on survival and growth? 3) Does microhabitat affect survival and growth?

Four cages, each containing twelve buckets with a single lobster per bucket, were deployed as a group with approximately 3 m spacing between adjacent cages. Four groups (N = 16 cages) were placed on soft bottom (mud, gravel, or sand depending on the location), and four groups were placed on hard bottom at each of the six sites. Cage one in each group contained Squat buckets with a substrate of moderately crushed soft-shell clam (*Mya arenaria*) shells that covered the bottom of the bucket to a depth of 3 cm (Fig. 4). Cage two contained Tall buckets without the additional substrate. Cage three contained Squat buckets without substrate, and cage four contained Tall buckets with a similar crushed shell substrate.

During October 2006, survival was estimated at each site (Table 2). Each group of four cages was hauled aboard the fishing vessel. One cage of the four was randomly chosen, and the presence or absence of a live juvenile lobster was noted. A second sampling occurred at five of six sites during the June 2007 (Table 2). All cages that could be located were brought aboard,

the contents of each bucket sieved through a 2 mm mesh, and the presence and absence of young lobsters similarly noted. The final sampling occurred approximately two years after the initiation date (Table 2). All animals were removed from all remaining buckets, and placed into numbered, plastic test tubes containing ambient seawater. Tubes were then placed into coolers (as described above) and taken to the Downeast Institute where the carapace length (CL; to the nearest 0.1 mm using Vernier calipers) and total mass (to the nearest 0.001g using an electronic balance) of each, after placing on paper toweling for 3 seconds, was recorded on the same day.

Statistical Analyses

Variances associated with the percent survival data for the June 2007 and August 2008 sampling dates were heterogeneous (Cochran's test for variance homogeneity, $P < 0.025$). Therefore, analysis of variance (ANOVA) was performed on the arcsine-transformed mean percent survival data. ANOVA was performed on the untransformed mean CL. The design at each site was a randomized complete block design (RCBD), with each block (four cages per line) in each habitat containing one replicate of each of the four treatments: 1) Squat buckets with shell; 2) Tall buckets without shell; 3) Squat buckets without shell; and, 4) Tall buckets with shell. The linear model was:

$$Y_{ijkl} = \mu + A_i + B_j + AB_{ij} + C(AB)_{k(ij)} + D_l + AD_{il} + BD_{jl} + ABD_{ijl} + DC(AB)_{kl(ij)}$$

Where:

Y_{ijkl} = dependent variable (arcsine-transformed percent survival, CL)

μ = theoretical mean;

A_i = Site (i = 6 sites along the Maine coast; factor is fixed)

B_j = Habitat (j = 2 habitats – hard- vs. soft-bottoms; factor is fixed)

C_k = Blocks (k = 4 blocks per habitat and site; factor is random)

D_l = Treatment (l = 4 treatments per block; factor is fixed)

Because this was a traditional RCBD (i.e., without replication of treatments within a block), sources of variation containing block effects cannot be tested; hence, only fixed factor sources of variation are shown with their respective P-values. When possible (balanced data), treatment

effects were decomposed into three, orthogonal single-degree-of-freedom sources of variation: 1) Bucket: Squat vs. Tall; 2) Microhabitat: Shell vs. No Shell; and, 3) Bucket x Microhabitat interaction. Similarly, the interaction between Treatment and Habitat was decomposed into three, orthogonal single-degree-of-freedom sources of variation: 1) Habitat x Bucket; 2) Habitat x Microhabitat; and, 3) Habitat x Bucket x Microhabitat.

Analysis of regression lines (testing for common slope) and subsequent analysis covariance (ANCOVA) was performed on the relationship between CL and total wet mass. Least square (adjusted) means were calculated, along with their standard errors, to examine spatial variation in the weight-length relationship.

Additional tests

I. Effect of cobble substrate on lobster growth and survival

To determine if lobster growth and survival varied as a function of cobble habitat, an additional study was initiated at the Beals Island study site on 5 August 2006. In this trial, 24 cages (eight pairs of three cages each) were deployed. Six 2-gallon (7.6 liter) plastic pails (top diameter = 20.8 cm, bottom diameter = 24.6 cm; constructed of food grade high density polyethylene) were added to each cage. The bottom of each pail was cut out leaving a 1 cm rim to which a piece of nylon window screening was hot-glued (as described above) forming a secure bottom. Two of the pails contained shell substrate (as described above) to a depth of 3 cm, two contained no substrate, and two contained 6-8 large rock cobble (diameter = 10-15 cm). For these last two pails, a circular insert constructed of vinyl-coated trap wire was placed over the nylon window screening in the bottom of the pail to hold up the cobble and ensure that it would remain in place and not puncture the screening. A large hole (18 cm diameter) was cut in the lid of each pail and a piece of nylon window screening was placed over the top of the pail and the lid snapped over the top of the pail securing the top screening. Cages were placed on hard bottom in the vicinity of the cages deployed a month before. One pair of three cages was sampled on 5 October 2006. Attempts were made to sample all cages on 11 August 2008.

II. Effect of compartment size on lobster growth – 2 August 2006 to 2 August 2007

Because lobsters were restricted to shelters (plastic buckets or pails) in this study, growth rates may have been affected by the size of the container (sensu McLeese 1972; Van Olst and Carlberg 1978). To examine potential effects of shelter size on lobster growth, a study was initiated on 2 August 2006 at Mud Hole Cove, Beals, Maine (44°29.14'N; 67°35.18'W). Ten wooden trays 122 cm x 91 cm x 7.6 cm deep were constructed and a series of ten compartments (two replicates of five different sizes) created in each using wooden strapping. Size of compartments was as follows: 1) 7.6 cm x 21.5 cm; 2) 18 cm x 22 cm; 3) 21.5 cm x 28 cm; 4) 44.5 x 28 cm; and, 5) 58 cm x 45 cm. The bottom of the tray was lined with nylon window screening (aperture = 1.8 mm). One stage IV-V cultured lobster (as described above) was placed into each compartment within one tray (N = 10), and a piece of window screening affixed to the top of the cage securing the animal within each compartment. Trays floated on the surface of Mud Hole Cove for one year. On 2 August 2007, trays were retrieved, and taken to the Downeast Institute, where the CL and wet mass (as described above) of each live lobster recorded as well as the size of compartment. A regression of CL (mm) vs. compartment area (cm^2) and mass (g) vs. container area was developed to determine if the slope of the line was significantly different from zero (H_o: Slope of the line = 0, indicating no relationship between the two variables).

III. Effect of container size on lobster growth and survival – 2 August 2006 to 19 November 2007

A second field test was conducted to examine effect of container size on fate and growth of cultured lobsters from 2 August 2006 to 19 November 2007 at Mud Hole Cove, Beals, Maine. Ten commercial "lantern nets" (frame size = 50 cm; 10 tiers; aperture = 15 mm) comprised of UV-resistant polyethylene material were used. The two bottommost tiers were not used in this trial. On tier 1 (top) and 8 (bottom), a single cultured lobster juvenile was added to each of three 15 cm diameter x 2.5 cm deep Petri dishes. Dishes were sandwiched between two pieces of vinyl-coated lobster trap wire (48 cm long x 20 cm wide) that were cinched together using nylon cable ties. Each Petri dish had an 83 mm hole drilled in both the bottom and cover that were

filled by placing a piece of nylon window screening (as described above) and affixing it to the plastic using PVC cement. Lobsters were added to Tall buckets (as described above) on tiers 2, 4, and 6. Two of the four buckets on each tier contained crushed shell substrate (as described above), and two contained no microhabitat. Squat buckets (as described above) were placed on tiers 3, 5, and 7. As with the Tall buckets, two of the Squat buckets on each tier contained a shell substrate, and two did not. Each net was independently anchored to the bottom using a cinder block filled with cement. A 15 cm diameter x 45 cm Styrofoam buoy was tied to the top of each net to help keep the net and its contents upright in the water column. A 3 m piece of rope was tied to the buoy and a similar size buoy attached to the other end of the rope, which served as a means of marking each net. Nets were deployed in 5 m of water at low tide. One of the ten nets was sampled on 2 August 2007 and taken to the Downeast Institute (DEI) where the CL and wet mass of all live animals measured (as described above). The remaining nine nets were sampled on 19 November 2007, and similar measurements were recorded for each living lobster.

Analysis of variance was performed on the untransformed, mean percent survival data (19 November 2007) using the following linear model:

$$Y_{ijklm} = A_i + B_j + AB_{ij} + C(B)_{k(i)} + AC(B)_{ik(j)} + D(CB)_{l(jk)} + AD(CB)_{il(jk)} + e_{m(ijkl)}$$

Where:

Y_{ijklm} = dependent variable (percent survival, CL)

μ = theoretical mean;

A_i = Net (i = 9 lantern nets; factor is random)

B_j = Container (j = 3 sizes – dishes, Squat buckets, Tall buckets; factor is fixed)

C_k = Tier (k = 2 (for dishes) or 3 (for buckets); factor is fixed)

D_l = Substrate (l = 2 – crushed shell vs. no shell; factor is fixed)

e_m = Experimental error

Analysis of the regression lines and ANCOVA were performed on the log-transformed wet mass vs. CL to examine effects of container size and substrate. A decision rule of $\alpha = 0.05$ was used for all statistical tests. Unless otherwise stated, untransformed means are given along with their 95% confidence intervals.

Results

October 2006 sampling

All cages at each site were recovered. Survival in the randomly sampled cage from each block varied from 53.1 ± 20.7% (Tenants Harbor, soft-bottom) to 93.8 ± 9.7% (Beals, hard-bottom) with an overall rate of 62.5 ± 9.7% (Table 3; Fig. 5). Although no lobsters were measured at that time, their color was noted and was distinctly darker and redder than animals appeared when the trial was initiated in July 2006 (Fig. 6).

Cages assigned to soft-bottoms had a tendency to collect sand and/or mud (Fig. 7), compared to those assigned to hard bottoms. This was especially evident at Beals, Tenants Harbor, and York, where some of the buckets were nearly completely filled with sediment. No association between microhabitat and amount of sediment in the buckets was observed. Sediment did not appear to affect lobster survival during this sampling period.

June 2007 sampling

Due to storms over the winter and spring, some gear was lost and/or buckets had filled with so much sediment that it was not possible to lift the gear on board the fishing vessel (Table 4; Fig. 8). The only exception was in Cutler Harbor, where all gear was retrieved. Most gear loss occurred at Boothbay Harbor and York during a coastal storm between 15-19 April 2007. The storm coincided with astronomically high tides, high winds, and flooding rivers (Kesich 2007). Maximum gust speed of wind recorded at the Maine Department of Marine Resources laboratory in West Boothbay Harbor on 16 April 2007 was 93.5 kph. Four U.S. Coast Guard navigational buoys near Boothbay Harbor were lost during that storm (A. Kenney, Boothbay Harbor, pers. comm.), and, as many as 67 buoys were dislodged or disabled from Maine to NewYork during that storm (http://www.provincetownbanner.com/article/banner_daily_update_article/_/47889/Banner_Daily_Update/4/26/2007). Sediment build-up in buckets occurred at all sites, but was especially severe at Beals and Boothbay Harbor (Table 4). For example, 92% and 94% of buckets in cages deployed on soft bottoms at these two sites were at least half-filled with mud,

coarse sand, or coarse shell. Ripped or torn screens or those with at least one large hole were observed at every site (Table 4), but the proportion of buckets with these problems was highest at Tenants Harbor (61.1% in hard bottoms) and York (41.7% in soft bottoms; 54.2% in hard bottoms).

Overall, mean survival was 46.5 ± 5.5% (n = 114); however, significant differences were observed between sites (P = 0.0035). A Student-Neuman-Keuls (SNK) test demonstrated that mean percent survival fell into two discrete groupings with Cutler and Tenants Harbor (64.0 ± 6.655%, n = 32; and, 59.3 ± 9.1%, n = 23, respectively) in one group, and the remaining three sites in the other group (32.0 ± 7.9%, n = 59). Habitat (soft- vs. hard-bottom) effects also were statistically significant (P = 0.0232), with overall lobster survival on soft-bottom habitats nearly 35% greater (54.6 ± 6.9%, n = 64) than on hard bottoms (36.1 ± 8.3%, n = 50) (Fig. 9). No significant effects due to treatment (bucket size, microhabitat, and their interaction) were detected (Table 5). At Cutler, the only site where all gear was recovered, and where a low percentage of the window screening was lost or damaged (ca. 1%), the same pattern of higher survival in soft bottoms (73.8 ± 11.8%, n = 16) vs. hard bottoms (54.2 ± 9.7%, n = 16) (Table 6) was observed.

August 2008 (final) sampling

A total of 76 (of 192, or 39.6%) cages were recovered across all sites (Table 7). Except for the Cutler study site, cages and buckets at most sites had apparently been tossed about as plastic lids and window screening covers had come off (Fig. 10) and were laying in the bottom of the cages. Some of the damage was due, again, to winter storms, but in several instances (Beals, Stonington, Tenants Harbor), fishermen reported that gear had been dragged by commercial sea scallopers and/or urchin harvesters. At the Beals study site, cages were so heavy (presumably because the experimental units [buckets] had filled with sediments) that the rope attached to the lead cage parted on five of the blocks, and all of the cages (n = 20) were lost.

A total of 338 lobsters were recovered from a total of 2,304 individuals initially deployed in July 2006 (14.6%). However, because only 76 cages were retrieved, the percent of lobsters

recovered is estimated to be 37.1% (Table 7). This is a conservative surrogate for percent survival because recovery rates were based on twelve undamaged buckets (and their screens) per cage (see Table 4 for data collected in 2007 on the status of these experimental units).

Mean percent of lobsters recovered was site specific ($P = 0.0011$; Fig. 11), but no overall effect of habitat or treatment was detected ($P > 0.25$; Table 8). The tests for habitat and treatment were not very powerful (Power < 0.55), presumably due to the loss of gear (replicates) which resulted in high variability between replicates within a treatment. Highest mean percent recovered occurred at Cutler (51.2 ± 8.2%, n = 27), and lowest at York (12.0 ± 4.7%, n = 9). An a posteriori SNK test was unable to separate the means unambiguously. Mean CL varied significantly across sites and treatments ($P < 0.001$); however, the interaction of these factors was also significant ($P < 0.0001$; Table 9; Fig. 12). Therefore, separate one-way ANOVA's and subsequent SNK tests were performed on the mean CL data for each site to determine specific treatment effects. Only two sites, Cutler and Tenants Harbor, showed significant treatment effects ($P < 0.025$; Fig. 12), and at each, lobsters were significantly larger in Squat buckets with shell vs. Tall buckets with shell (by 8.7% [Cutler] and 25.8% [Tenants Harbor]). At both sites, no significant difference was observed in mean CL between animals housed in Squat or Tall buckets without microhabitat. Similarly, a separate one-way ANOVA was performed on the mean CL data to examine spatial variation. Significant variation in mean CL was observed between sites ($F = 21.28$, df = 5, 70, $P < 0.0001$), and SNK revealed that lobsters at Beals and Stonington attained similar sizes (mean CL = 14.3 ± 0.77 mm, n = 15 cages) that were significantly greater by nearly 35% than lobsters from the remaining sites (mean CL = 10.7 ± 0.34 mm, n = 61 cages; Table 10; Fig. 13). In addition, size-frequency distributions for animals recovered from each site were significantly different (6 x 5 G-test of independence; $G = 188.5$, df = 20, $P < 0.001$; Fig. 14).

The relationship between wet mass and CL was allometric ($r^2 = 0.975$; Fig. 15). Analysis of log-transformed regression lines relating weight and CL indicated that the slopes were equal ($F = 0.24$, df = 5, 321, $P = 0.9469$), and analysis of covariance demonstrated a significant effect due to site ($F = 8.45$, df = 5, 326, $P < 0.0001$). Least square (adjusted) means for wet mass (using a grand mean CL of 11.14 mm) indicated that lobsters from Boothbay Harbor and Cutler had

lower weights for a given CL than animals from Tenants Harbor, York, or Stonington. Animals from Beals were similar to both groups (Fig. 16).

Additional tests

I. Effect of cobble substrate on lobster growth and survival

One block of three cages containing the 2-gallon pails (2 pails with cobble, 2 pails with shell substrate, 2 pails without substrate) was sampled on 5 October 2006. All lobsters (N = 18) were found alive. Similarly, one block of three cages was sampled on 15 June 2007. One cage contained no live lobsters as the window screening was ripped or torn on each. In the second cage, both lobsters were alive in the two pails with and without shell, and one lobster was alive in one of the pails with cobble (the window screening was torn in the second pail). In the third cage, both lobsters were alive in the pails with shell, one of two lobsters were alive in the pails without substrate, and the screening was torn on the other two pails with cobble substrate. On 11 August 2008, five of the eight blocks of cages were missing (none of the buoys marking the blocks could be found at low tide). The remaining three blocks of three cages were lost during the hauling when the rope parted on each.

II. Effect of compartment size on lobster growth – 2 August 2006 to 2 August 2007

Seven of the ten trays suffered damage from winter ice that built up in Mud Hole Cove in January 2007. The structural integrity of these trays was compromised, and no live lobsters were recovered from them. The remaining three trays also suffered some damage from ice, but not as severe, as a total of seven animals were recovered from these trays ($n_A = 5$; $n_B = 1$; $n_C = 1$). Survival was not estimated as many of the compartments of these trays also had large holes in the bottom screening. There was a positive, linear trend relating CL and mass to compartment area (Fig. 17), and the slope of both relationships was significantly greater than zero ($P < 0.05$) suggesting that both carapace length and mass increase significantly with increasing compartment size.

III. Effect of container size on lobster growth and survival – 2 August 2006 to 19 November 2007

Survival in the lantern nets was low (26.6% in August 2007, n = 1; 24.1 ± 6.1% in November 2007, n = 9). All nets had become heavily fouled with macroalgae (*Laminaria longicruris*) and many were resting on the bottom. None of the mesh screening on any experimental units was ripped or torn; however, > 50% were filled with soft mud. It is unknown whether the mud contributed to lobster mortality or appeared after the lobsters died. Most of the containers with live lobsters, independent of container size, had little or no mud indicating active movement of the animal inside. Animals sampled from the net in August 2007 had a mean CL of 11.2 ± 0.8 mm (n = 8). Lobsters in the remaining nine nets were 12.5% larger in November 2007 (12.6 ± 0.5 mm, n = 65).

No significant variability in lobster survival was observed between nets (P = 0.3662), nor was there an overall effect due to container size (P = 0.9802); however, the effect of container size varied significantly from net-to-net (P = 0.0212; Table 11). Separate one-way ANOVA's were performed to further examine this interaction source of variation; however, no pattern was revealed. For example, container size was a significant source of variation for three of the nine nets but none of the tests had the same results (Net 3: survival in the dishes was greater than the combined mean of the Squat and Tall buckets – 66.7% vs. 12.5%; Net 3: survival in Squat buckets was lower than in Tall buckets – 8.3% vs. 50%; Net 6: survival in Squat buckets was greater than in Tall buckets – 41.7% vs. 8.3%). Lobster survival was approximately 20% greater in buckets with shell vs. those without (25.9 ± 8.3% vs. 8.2%).

Container size had a significant effect (P = 0.011) on mean CL (Table 12), and was 33% greater for lobsters in buckets (regardless of substrate; 13.3 ± 0.52 mm, n = 51) vs. those in the dishes (10.0 ± 0.53, n = 14). Analysis of regression lines (log-transformed wet weight vs. CL) demonstrated that the slopes of the lines were similar (F = 1.04, df = 1, 55; P = 0.3116) and ANCOVA showed that there was no significant effect due either to container size (F = 2.06, df = 2, 60; P = 0.1360) or substrate (F = 3.05, df = 1, 60; P = 0.0859).

Discussion

This study assessed regional variation in growth and survival of juvenile American lobsters, *Homarus americanus*, along the Maine coast over a two-year period. Crustaceans lack specific visible age markers which makes determining age of wild animals difficult. Although techniques using the fluorescent aging pigment lipofuscin (Sheehy 1990; Wahle et al. 1996; Tully et al. 2000; Uglem et al. 2005) may produce accurate estimates of age of wild animals, especially when the temperature regime under which animals live is known, an unambiguous method to age animals is to begin with animals that are of known age. Here, cultured lobsters (stage IV-V) were used to assess effects of habitat (soft- vs. hard-bottoms) and microhabitat (shell vs. no shell) on growth and survival of lobsters in flow-through containers of varying sizes at six study sites from York (southernmost) to Cutler (northernmost).

This study utilized a new methodology for holding lobsters in the field (Beal et al. 2002). An individual lobster entrapped in a flow-through container is able to survive and grow by feeding on detrital particles entering the container through the apertures of the window screening, or from organisms that settle into and grow on the hard surfaces of the container as invertebrate larvae or spores of macroalgae. Recent work in eastern Maine has shown that if containers are arrayed in the water column, survival rates can be as high as ca. 90% over an 11-month period (Beal 2006). In the present study, containers were placed within larger cages (that were grouped into a block of four cages) and deployed on the bottom in both soft- and hard-bottom habitats at each site. Number of animals recovered in the present study was low (338 of 2,304 deployed, or ca. 15%), presumably the result of natural mortality as well as from storm and other events that resulted in blocks of cages moving 100's of meters from where they were deployed as well as containers filling with sediments and/or fine shell fragments (Fig. 8). Surprisingly, in some instances (especially at York and Tenants Harbor), containers were completely filled with soft sediments yet some lobsters within were alive.

Recovery rates were highest at the site that was physically the most protected (Cutler Harbor, ca. 50%), and lowest where gear had moved the most (Boothbay Harbor and York – Table 7). This undoubtedly contributed to the significant effect of site on mean percent recovered, and likely

influenced growth rates (see below). Because juvenile lobsters inhabit many different hard and soft bottoms (Cooper et al. 1975; Elner and Hamet 1984; Cooper and Uzmann 1980; Able et al. 1988), this study had the potential to answer whether habitat (soft- vs. hard-bottom) significantly affected lobster survival. Although the ANOVA (Table 8) on final mean recovery rate showed that habitat was not a statistically significant source of variation, the high rate of gear loss resulted in low statistical power, even though mean recovery on hard bottoms was approximately 35% higher than from soft bottoms (45.6 ± 9.9%, n = 21 vs. 33.6 ± 5.5, n = 55). For example, during the June 2007 sampling, when every cage from five of six sites that could be was sampled (114 of 160, or 71%) the effect of habitat was statistically significant (Table 5). Lobster recovery rate was 52% greater from cages deployed over soft bottoms (54.6 ± 6.9%, n = 64) than over hard bottoms (36.1 ± 8.3%, n = 50). I examined results from June 2007 and August 2008 from the single site, Cutler, with the most complete data. In 2007, 73.8 ± 6.8% of animals from soft bottoms were recovered compared to 54.2 ± 9.7% (n = 16) from hard bottoms (P = 0.0380). In 2008, no significant habitat differences were detected (P = 0.0828), although there was a 12% higher recovery rate from cages on hard vs. soft bottoms (53.8 ± 10.6%, n = 15 vs. 47.9 ± 14.7%, n = 12). This indicates essentially no losses of lobsters from hard bottoms over the 14-month period, but a 54% reduction on soft bottoms over that time.

Absolute growth (measured as final mean CL) varied significantly across sites (P = 0.0002; Table 9), but did not yield results that were anticipated. Seawater temperatures along the coast of Maine generally increase in a southwesterly direction due to the weakening influence of the Labrador Current (Wanamaker et al. 2008). That is, surface and subsurface waters are generally cooler in eastern Maine and become progressively warmer towards the Maine/New Hampshire border. This general phenomenon is confirmed by temperature probes deployed on commercial lobster traps located in the coastal waters along the Maine coast (http://www.emolt.org/). Therefore, final mean CL was expected to be smallest in eastern Maine (Beals and Cutler) and greatest in southwestern Maine (Boothbay Harbor and York) with intermediate sizes expected in Stonington and Tenants Harbor. However, final mean CL groupings from ANOVA and SNK-tests demonstrated that animals from Beals and Stonington (14.4 ± 0.59 mm, n = 53) were nearly 35% larger than those from the other four sites (10.7 ± 0.17 mm, n = 284) (Table 10; Figs. 12, 14). Besides possible variation in food supply between sites (not measured in this study), final

mean size could be a function of stress due to movement of gear, percent of containers filled with coarse or fine sediments, or other unknown causes. Although the same experimental design was employed at each site, the actual on-bottom conditions that lobsters were exposed to was very different between sites. For example, what one fisherman may have called "soft" bottom vs. "hard" bottom may have differed based availability of habitat. Soft bottoms can be defined by some as either sandy, gravelly, or muddy. In addition, water depths were not constant between sites. One location in Boothbay Harbor (0.3 km southwest of the White Island), where the highest percent recovery for that site was noted, was the deepest of all sites (37 m). Animals at that deep water site were smaller than those collected from shallower (2-5 m) areas in that same region (Fig. 18). Final mean CL, then, was not only a function of geographic location along the Maine coast, but other factors that were not consistent between sites.

Size of container had a significant effect on final mean CL at two of the six sites, but only when shell was used as a microhabitat. Animals housed in "Squat" buckets at Cutler and Tenants Harbor had 8.7% and 25.8% greater final mean CLs, respectively, than lobsters in Tall buckets. Squat buckets had a 34% greater surface area than Tall buckets, but had smaller volumes (by nearly 1 L). Two additional attempts were made to determine the relationship between container/compartment size and juvenile lobster growth. Animals were housed in flow-through containers placed into submerged lantern nets and in flow-through compartments that floated on the surface of the water at a small cove in eastern Maine (Mud Hole Cove, Beals). Although survival was poor in both experiments for different reasons, results generally indicated that animals grew larger when offered more space. Lobster juveniles were 33% larger in buckets vs. Petri dishes (lantern net experiment) at the end of 15 months, and there was a significant trend for animals to grow larger when offered larger compartments over a 12-month period (Fig. 17). These results beg the question whether or not the observed final mean CLs across the six study sites are representative of wild two year-old lobsters, or was growth diminished because animals were shelter-restricted, may have been food-limited, or were stressed in other ways that had a negative effect on growth?

Growth estimates from the laboratory (Templeman 1948; Hughes et al. 1972; Barshaw and Bryant-Rich 1986) and cohort analyses based on size-frequency distributions of wild-caught

juveniles (Hudon 1987) suggest that lobsters as large as 17 mm CL are in their first year. Gendron and Sainte-Marie (2006) examined size-frequencies of lobster juveniles around Îles de la Madeleine (Quebec, Canada). They showed that the size of first-year animals was highly variable, ranging from 4.0 to 13.5 mm CL, and that mean CL after the first year was 10.0-10.5 mm. After two years, animals attained lengths of approximately 20-25 mm CL. Cowan et al. (2001) tagged and followed individuals as small as 12 mm CL collected from the intertidal near Orr's Island, Harpswell, Maine. They discovered that growth was seasonal, occurring between April and November, and that growth was approximately linear. They concluded that juveniles may reach sizes up to approximately 20 mm CL in their second year. Overall mean CL in the present study was 11. 3 ± 0.22 mm (n = 337) with a range from 5.9 to 19.1 mm. Since average initial size was 4.1 mm CL, the range of CLs represents an increase between 44% and 365% over two years.

Because this was the first attempt to follow the fate and growth of known-age animals from stage IV-V in the field without imposing tags or other markers on/within the animals, which may influence their behavior and growth (Comeau and Savoie 2001), it is difficult to know whether the growth estimates observed here are representative of the population of wild juveniles of comparable sizes. Whether methodological choices biased results in this study remains to be seen. Future field investigations to determine growth-at-age should examine effects of: 1) container size/shape over a wider range of sizes (see Van Olst and Carlberg 1978); 2) water depth within a given location; 3) habitat stability within a given location; and 4) initial size on growth trajectory.

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Table 1. Initial mean size (carapace length, total length) and wet mass (g) (± upper and lower 95 % Confidence Limits) of 36 cultured individuals of the American lobster, *Homarus americanus*, from a representative sample of animals taken on 28 July 2006 at the Downeast Institute for Applied Marine Research & Education (Beals, Maine). Once animals reached stage IV in communal culture tanks, they were transferred to plastic, individual compartments (65 mm x 65 mm x 70 mm), held at ambient seawater conditions, and fed cultured brine shrimp until they were used in field trials.

Measured attribute	Mean	Lower 95% CL	Upper 95% CL
Carapace Length (mm)	4.14	3.99	4.29
Total length (mm)	18.10	17.30	18.91
Wet mass (g)	0.065	0.057	0.072

Table 2. Geographic distribution of study sites along the Maine coast, approximate latitude and longitude of the study site, initiation date, and sampling dates associated with the juvenile lobster survival/growth trial. Initiation and first sampling occurred in 2006, second sampling in 2007, and final sampling in 2008.

Site	Latitude/ Longitude	Depth (m)	Initiation Date	First Sampling	Second Sampling	Final Sampling
Cutler	44°39.11'N 67°12.15'W	2-5	23 July	18 October	20 June	7 August
Beals	44°28.06'N 67°37.17'W	2-5	12 July	5 October	15 June	11 August
Stonington	44°01.85'N 68°35.28'W	12-20	16 July	8 October	nd	24 August
Tenants Harbor	44°58.26'N 67°69.11'W	10-20	27 July	13 October	21 June	14 August
Boothbay Harbor	44°47.64'N 69°34.56'W	3-37	29 July	15 October	27 June	13 August
York	43°07.80'N 70°36.87'W	10-20	26 July	19 October	22 June	28 August

Table 3. Mean percent survival (± 95% CI) of juvenile lobsters at each site during the October 2006 sampling (see Table 2 for specific dates). One cage per block was sampled.

Site	Date	Bottom Type	% Survival ± 95% Confidence Limits
Cutler	18 October	Hard	59.38 ± 29.43
		Soft	56.25 ± 24.15
Beals	5 October	Hard	93.75 ± 9.68
		Soft	75.00 ± 25.01
Stonington	8 October	Hard	62.50 ± 19.35
		Soft	53.13 ± 17.44
Tenants Harbor	13 October	Hard	50.00 ± 30.00
		Soft	53.13 ± 20.71
Boothbay Harbor	15 October	Hard	57.50 ± 23.54
		Soft	56.25 ± 24.35
York	19 October	Hard	59.71 ± 11.23
		Soft	73.96 ± 16.42

Table 4. Status of gear, proportion of buckets filled with mud, and percent survival (± 95% CI) of juvenile lobsters at each study site in June 2007 (see Table 2 for specific sampling dates). A total of twelve buckets (with a single juvenile lobster) was added to each submerged cage. Sample size for mean percent survival ranged from 1 to 4 depending on the number of cages recovered and buckets that were capable of containing/retaining lobster juveniles.

Site	Habitat	No. Cages Recovered	Buckets[1] Screening	Buckets[2] Sediment	Mean Percent Survival			
					Squat Buckets		Tall Buckets	
					With Microhabitat	Without	With Microhabitat	Without
Cutler	Soft	16	1	12	85.4(12.7)	72.9(16.7)	72.9(12.7)	64.2(26.5)
	Hard	16	2	0	58.3(44.6)	56.3(33.2)	45.8(17.1)	56.3(22.7)
Beals	Soft	6	6	66	7.4(15.9)	25.0	0.0(-[3])	0.0(-)
	Hard	12	10	24	70.1(16.6)	44.4(97.8)	43.9(98.7)	38.9(99.5)
Tenants	Soft	16	8	22	62.5(13.3)	44.3(21.1)	68.4(36.1)	59.3(22.1)
Harbor	Hard	9	66	0	16.7(-)	50.0(0.0)	100.0(-)	69.8(71.3)
Boothbay	Soft	4	3	45	18.1(88.2)	45.8(264)	12.5(159)	29.2(371)
Harbor	Hard	12	4	36	16.7(106)	30.9(30.3)	45.5(125)	25.0(318)
York	Soft	12	60	20	44.4(104)	46.0(99.2)	46.8(30.3)	54.2(125)
	Hard	12	78	56	16.7(71.7)	8.3(35.8)	20.0(49.7)	15.6(37.3)

[1] Number with missing top screens or lids, or holes in screening top or bottom
[2] Number that were at least half-filled with sediment (mud or coarse sand or shell)
[3] n = 1

Table 5. Analysis of variance on the arcsine-transformed mean percent survival data of juvenile lobsters from the June 2007 sampling at five of the six sites (see Table 2 for specific sites and sampling dates). Treatment refers to four fixed, orthogonal factors ("Squat" and "Tall" buckets with or without shell as a microhabitat). Habitat refers to submerged cages deployed on soft or hard bottom at each study site. Four cages containing twelve buckets each were deployed in four blocks within each habitat at each site in July-August 2006. Because gear was lost or damaged, missing values resulted in an unbalanced data set; therefore, Type II sums of squares were used for all hypothesis tests (see Langsrud 2003).

Source of variation	df	SS	MS	F	Pr>F
Site	4	14782.75	3695.59	5.41	0.0035
Habitat	1	4068.14	4068.14	5.95	0.0232
Site x Habitat	4	4657.15	1164.29	1.70	0.1852
Treatment	3	314.94	104.97	0.35	0.7911
Site x Treatment	12	2811.62	234.30	0.78	0.6722
Habitat x Treatment	3	488.87	162.96	0.54	0.6574
Site x Habitat x Treatment	12	2121.57	176.79	0.59	0.8440
Block(Site x Habitat)	22	15039.25	683.60	-	No test
Treatment x Block(St x Hab)	52	15708.41	302.08	-	No test

Table 6. Analysis of variance on the arcsine-transformed mean percent lobster survival data from Cutler on 18 October 2007. Juveniles (stages IV-V), cultured at the Downeast Institute for Applied Marine Research and Education, were placed individually into buckets (3.3 L or 4.2 L) that permitted seawater to flow in and out, but retained the lobsters. Buckets (n = 12) were placed into wire cages that were placed on either soft or hard bottom within Cutler Harbor on 23 July 2006. Cages were arrayed in blocks of four with a single treatment assigned randomly to each cage within the block. Treatments were a factorial combination of bucket volume and microhabitat (crushed soft-shell clam shells vs. no shells). Orthogonal, single degree-of-freedom tests appear below each fixed source of variation with greater than 1 df.

Source of variation	df	SS	MS	F	Pr>F
Habitat	1	1176.11	1176.11	7.03	0.0380
Treatment	3	382.11	127.37	1.57	0.2310
Bucket	1	262.04	262.04	3.23	0.0890
Microhabitat	1	71.27	71.27	0.88	0.3609
Bucket x Microhabitat	1	48.80	48.80	0.60	0.4479
Habitat x Treatment	3	201.89	67.29	0.83	0.4945
Sft v. Hard x 3.2L v. 4.2L	1	18.38	18.38	0.23	0.6397
Sft v. Hard x Shell v. Noshel	1	7.69	7.69	0.09	0.7615
Sft v. Hard x Buck x Micro	1	175.82	175.82	2.17	0.1581
Block(Site x Habitat)	6	1003.77	167.29	-	No test
Treatment x Block(St x Hab)	18	1459.27	81.07	-	No test

Table 7. Location of each block of recovered cages, number of cages recovered, total number and percent of lobsters recovered at each site during the August 2008 sampling (see Table 2 for specific dates for each site). Total cages deployed = 192. Total number of lobsters deployed = 2,304. Percent recovered is the percentage recovered from cages recovered not cages deployed.

Site	Habitat	Block	Latitude	Longitude	No. Cages	Total No. Lobsters Recovered	Percent Recovered
Cutler	Hard	1	44°38.98'N	67°11.89'W	3	14	38.8
	Hard	2	44°39.11'N	67°12.22'W	4	18	37.5
	Hard	3	44°39.21'N	67°11.84'W	4	34	70.8
	Hard	4	44°39.30'N	67°12.12'W	4	31	64.6
	Soft	1	44°39.09'N	67°12.00'W	4	26	54.2
	Soft	2	44°39.12'N	67°12.18'W	4	19	39.5
	Soft	3	44°39.28'N	67°12.15'W	4	24	50.0
Beals	Soft	1	44°27.86'N	67°36.83'W	3	7	19.4
	Soft	2	44°27.88'N	67°36.86'W	2	9	37.5
	Soft	3	44°27.91'N	67°36.94'W	1	1	8.3
Stonington	Hard	1	44°01.85'N	68°35.82'W	2	9	37.5
	Soft	1	44°01.56'N	68°36.05'W	4	16	33.3
	Soft	2	44°01.60'N	68°36.07'W	3	11	30.5
Tenants	Hard	1	43°57.75'N	69°10.21'W	2	4	16.7
Harbor	Hard	2	43°58.27'N	69°10.21'W	1	2	16.7
	Soft	1	43°57.76'N	69°09.84'W	4	20	41.7
	Soft	2	43°57.77'N	69°09.91'W	4	23	47.9
	Soft	3	43°57.86'N	69°11.87'W	3	16	44.4
	Soft	4	43°57.91'N	69°11.85'W	4	21	43.8
Boothbay	Soft	1	43°47.03'N	69°34.57'W	3	12	33.3
Harbor	Soft	2	43°47.64'N	69°34.56'W	1	1	8.3
	Soft	3	43°49.57'N	69°35.05'W	3	7	19.4
York	Hard	1	43°07.80'N	70°36.87'W	1	3	25.0
	Soft	1	43°07.21'N	70°37.36'W	3	3	8.3
	Soft	2	43°07.23'N	70°37.26'W	2	2	8.3
	Soft	1	43°07.27'N	70°37.36'W	3	5	13.9
				TOTAL	76	338	37.1

Table 8. Analysis of variance on the arcsine-transformed mean percent lobster recovery data from all study sites in August 2008. Gear loss due to storms and other circumstances resulted in an unbalanced data set. Type II sums of squares were used to test all hypotheses.

Source of variation	df	SS	MS	F	Pr>F
Site	5	4687.49	937.49	7.36	0.0011
Habitat	1	4.02	4.02	0.03	0.8613
Site x Habitat	3	1053.82	351.27	2.76	0.0787
Treatment	3	396.35	132.12	1.22	0.3189
Site x Treatment	13	1438.74	110.67	1.02	0.4555
Habitat x Treatment	3	256.62	85.54	0.79	0.5082
Site x Habitat x Treatment	2	291.66	145.83	1.35	0.2750
Block(Site x Habitat)	15	1910.53	127.37	-	No test
Treatment x Block(St x Hab)	29	3131.73	107.99	-	No test

Table 9. Analysis of variance on the untransformed mean carapace length of live, juvenile lobsters recovered from experimental units at each site in August 2008 (see Table 2 for specific sampling dates). Gear loss and lobster mortality resulted in an unbalanced data set; hence, Type II sums of squares are used for all hypothesis tests.

Source of variation	df	SS	MS	F	Pr>F
Site	5	170.28	34.06	10.34	0.0002
Habitat	1	1.92	1.92	0.58	0.4571
Site x Habitat	3	4.14	1.38	0.42	0.7417
Treatment	3	5.98	1.99	7.56	0.0007
Site x Treatment	13	21.30	1.64	6.21	<0.0001
Habitat x Treatment	3	4.54	1.51	5.74	0.0033
Site x Habitat x Treatment	2	1.49	0.75	2.83	0.0752
Block(Site x Habitat)	15	49.38	3.29	-	No test
Treatment x Block(St x Hab)	29	7.65	0.26	-	No test

Table 10. Mean (± 95% CL) carapace length (mm) and wet mass (g) for live juvenile lobsters recovered from experimental units ("Squat" and "Tall" buckets) from each study site in August 2008 (see Table 2 for specific sampling dates). Data is pooled across blocks, habitat (soft- vs. hard-bottom), and treatments.

Site	n	CL	min	max	Lower	Upper	n	Mass	min	max	Lower	Upper
Cutler	166	10.4	7.0	14.2	10.2	10.6	165	0.65	0.17	1.81	0.61	0.69
Beals	17	14.8	5.9	19.1	13.3	16.2	17	2.29	0.10	5.02	1.74	2.85
Stonington	36	14.2	10.9	17.2	13.6	14.8	34	2.06	0.84	3.26	1.79	2.33
Tenants Harbor	85	11.7	7.9	15.7	11.3	12.1	85	1.05	0.24	2.51	0.94	1.16
Boothbay Harbor	20	10.0	7.5	13.3	9.1	10.9	20	0.63	0.22	1.47	0.43	0.81
York	13	10.6	9.0	12.8	9.8	11.4	13	0.75	0.40	1.35	0.56	0.92

Table 11. Analysis of variance on the mean percent survival of cultured lobsters held within each of three sizes of flow-through containers: 15 cm x 1 cm Petri dishes; 20 cm x 15 cm "Squat" plastic buckets; 17 cm x 20 cm "Tall" plastic buckets (described in methods section). Containers were arrayed within eight lantern nets from 2 August 2006 to 19 November 2007 at Mud Hole Cove (water depth at low tide ca. 5 m). A single cultured lobster (0_{CL} ± 95% CI = 4.2 ± 0.13 mm (range = 3.5 – 5.1 mm); 0_{Mass} ± 95% CI = 0.053 ± 0.005 g (range = 0.02 – 0.09 g) was added to each container. Three dishes were placed on the top (1st) and bottom (8th) tier of each net. Four Tall buckets were each added to tiers 2, 4, and 6, and four Squat buckets were each added to tiers 3, 5, and 7. Two buckets on each level contained no substrate, and two contained a coarsely crushed shell (*Mya arenaria*) substrate that covered the bottom of the container to a depth of 3 cm. Nets are considered a random factor, whereas level, container, and substrate are all fixed factors. *A priori* contrasts for Container appear below this source of variation.

Source of variation	df	SS	MS	F	Pr > F
Net	8	1.5185	0.1898	1.10	0.3662
Container	2	0.0648	0.0324	0.10	0.9802
Dish vs. Bucket	1	0.0231	0.0231	0.07	0.7959
Buckets: Tall vs. Squat	1	0.0417	0.0417	0.12	0.7287
Net x Container	16	5.3519	0.3344	1.94	0.0212
Tier(Container)	5	0.6296	0.1259	0.70	0.6272
Net x Tier(Container)	40	7.2037	0.1801	1.04	0.4132
Substrate(Tier, Container)	6	2.7500	0.4583	3.14	0.0111
Net x Subst(Tier, Container)	48	7.0000	0.1458	0.85	0.7453
Error	144	24.8333	0.1725		
Total	269	49.3519			

Table 12. Analysis of variance on mean CL of live lobsters held in lantern nets at Mud Hole Cove, Beals, Maine from 2 August 2006 to 19 November 2007. Lobsters were held in one of three different sized containers (Petri dishes; "Squat" and "Tall" buckets – see Table 11 and methods section for specific sizes). Each lantern net had ten tiers, but only the top nine were used. Lobsters used in the study were cultured at the Downeast Institute (Beals, Maine) and were stage IV-V ((0_{CL} ± 95% CI = 4.2 ± 0.13 mm (range = 3.5 – 5.1 mm); 0_{Mass} ± 95% CI = 0.053 ± 0.005 g (range = 0.02 – 0.09 g).

Source of variation	df	SS	MS	F	Pr > F
Net	8	34.09	4.26	2.70	0.0644
Container	2	83.66	41.83	12.07	0.0011
Dish vs. Bucket	1	83.10	83.10	18.11	<0.0001
Buckets: Tall vs. Squat	1	0.56	0.56	0.12	0.6578
Net x Container	13	46.35	3.57	2.26	0.0915
Tier(Container)	5	15.51	3.10	0.89	0.5097
Net x Tier(Container)	16	51.29	3.21	2.03	0.1179
Substrate(Tier, Container)	5	5.98	1.19	0.39	0.8363
Net x Subst(Tier, Container)	4	12.34	3.08	1.96	0.1709
Error	11	17.33	1.58		
Total	64	266.55			

Figure 1. a) Size-frequency distribution of juvenile, cultured American lobsters, *Homarus americanus*, (n = 36) used in the regional survival/growth investigation. b) Relationship between carapace length and total length for juvenile lobsters and 95% confidence limits ((Y = -1.47 + 4.724 X; r^2 = 0.8295; P < 0.0001; n = 36). c) Relationship between carapace length and wet weight for juvenile lobsters (Y = -0.117 + 0.043 X; r^2 = 0.8072; P < 0.0001; n = 36).

Figure 2. a) Photograph of the bottom of a "Tall bucket" used to contain individual lobsters. A hole (11.4 cm diameter) was cut in the bottom of each bucket, then a piece of nylon window screening was hot glued in place to cover the hole. b) Photograph of "Tall bucket" with lid and 23 cm x 23 cm piece of nylon window screening.

Figure 3. a) Photograph of a wire cage containing 12 "Squat buckets." b) Photograph of a wire cage containing 12 "Tall buckets." Each bucket was initially seeded with a single, culture stage IV/V lobster.

Figure 4. Photograph of "Squat buckets" with crushed soft-shell clam, *Mya arenaria*, shells as a microhabitat and additional surface area for fouling organisms to attach.

Figure 5. Mean percent survival (+ 95% CI) of juvenile lobsters from each of the six study sites. (See Table 2 for sampling dates during October 2006.) Each block of four cages in both hard- and soft-bottom habitats were hauled aboard the fishing vessel. One cage from each block was sampled at random and the presence or absence of live lobsters noted. (n = 4).

Figure 6. Photographs of lobsters taken during the first sampling in October 2006 from a) Beals; b) York; c) Tenants Harbor; and, d) Stonington. (See Table 2 for specific sampling dates.) Mesh size in photographs a) and c) is 2 mm. Window screening is seen in photograph d) (ca. 1.8 mm aperture).

Figure 7. Photographs of buckets with large amounts of sediments observed during the first sampling in October 2006 from a) and b) York; c) Tenants Harbor; and, d) Stonington. Each bucket contained a live lobster.

Figure 8. Photographs of buckets with large amounts of sediments (mud, coarse sand, coarse shell) during the second sampling in June 2007 from a) and b) Boothbay Harbor; c) York, and d) Beals. Live lobster juveniles are encircled with the yellow oval in b) and c).

Figure 9. Interaction plot demonstrating the relationship between mean percent survival and habitat across each site. ANOVA (Table 5) showed significant differences between sites and habitats with Cutler and Tenants Harbor having the highest mean survival and the other three sites having lower (and statistically similar) survival. Significantly higher overall juvenile lobster survival (ca. 35%) occurred

in cages deployed on soft vs. hard bottoms. (n varies from 6 to 16 cages per site and habitat).

Figure 10. Photographs taken in August 2008 of cages and buckets from a) Tenants Harbor and b) York. Many of the lids and nylon window screening of the buckets had come off due to movement of cages over the bottom during storms and, in several cases (Beals, Stonington, Tenants Harbor) due to being dragged over the winter by commercial urchin and/or scallop fishermen.

Figure 11. Mean percent of lobsters recovered (+ 95% CI) from cages in August 2008 from each study site (see Table 2 for specific sampling dates). Percent recovered was calculated by taking the number of live lobsters per cage and dividing by the number of buckets initially stocked per cage (n = 12). ANOVA indicated that percent recovered was site specific (P = 0.0011; Table 8); however, an a posteriori SNK test was unable to separate means unambiguously.

Figure 12. Interaction plot showing mean final carapace length of live lobsters recovered from each site across each of the four treatments. Squat and Tall refer to the size of the plastic, flow-through buckets (3.3 L vs. 4.2 L). Shell and No Shell refers to the microhabitat within buckets (crushed soft-shell clam shells vs. no microhabitat). ANOVA (Table 9.) indicated that both main and interactive effects were highly significant (P < 0.001).

Figure 13. Largest lobster recovered from experimental units at York on 28 August 2008. Carapace length = 12.8 mm; Wet weight = 1.345 g. Hand and lobster gauge belong to Pat White.

Figure 14. Initial (blue) and final size-frequency distribution of juvenile lobster carapace length (mm) from each study site. Lobsters were deployed at each site during July 2006 and collected in August 2008 (see Table 2 for specific dates for each year). Data from each site are pooled over habitat, bucket size, and microhabitat.

Figure 15. Allometric relationship between wet mass (g) and carapace length (mm) for live lobsters recovered from experimental units in August 2008 (see Table 2 for specific sampling dates). $Y = (0.000215)(X^{3.411})$, n = 333, $r^2 = 0.9746$.

Figure 16. Least square means for wet weight (g) ± SE for live lobsters recovered from experimental units in August 2008 from each study site (see Table 2 for specific sampling dates). Least square means are adjusted for a common CL of 11.1 mm.

Figure 17. Relationship between a) CL and container size ($Y = 11.3 + 0.002 X$; $r^2 = 0.774$, n = 7; P = 0.012) and b) Mass and container size ($Y = 0.663 + 0.00099 X$; $r^2 = 0.870$, n = 7; P = 0.002) from floating trays at Mud Hole Cove, Beals, Maine from 2 August 2006 to 2 August 2007.

Figure 18. Relationship between wet mass and CL for lobsters recovered from Boothbay Harbor on 13 August 2008. Animals recovered from one site at 37 m (deep) had smaller animals than those recovered from shallow sites (3-5 m = shallow).

Figure 1.

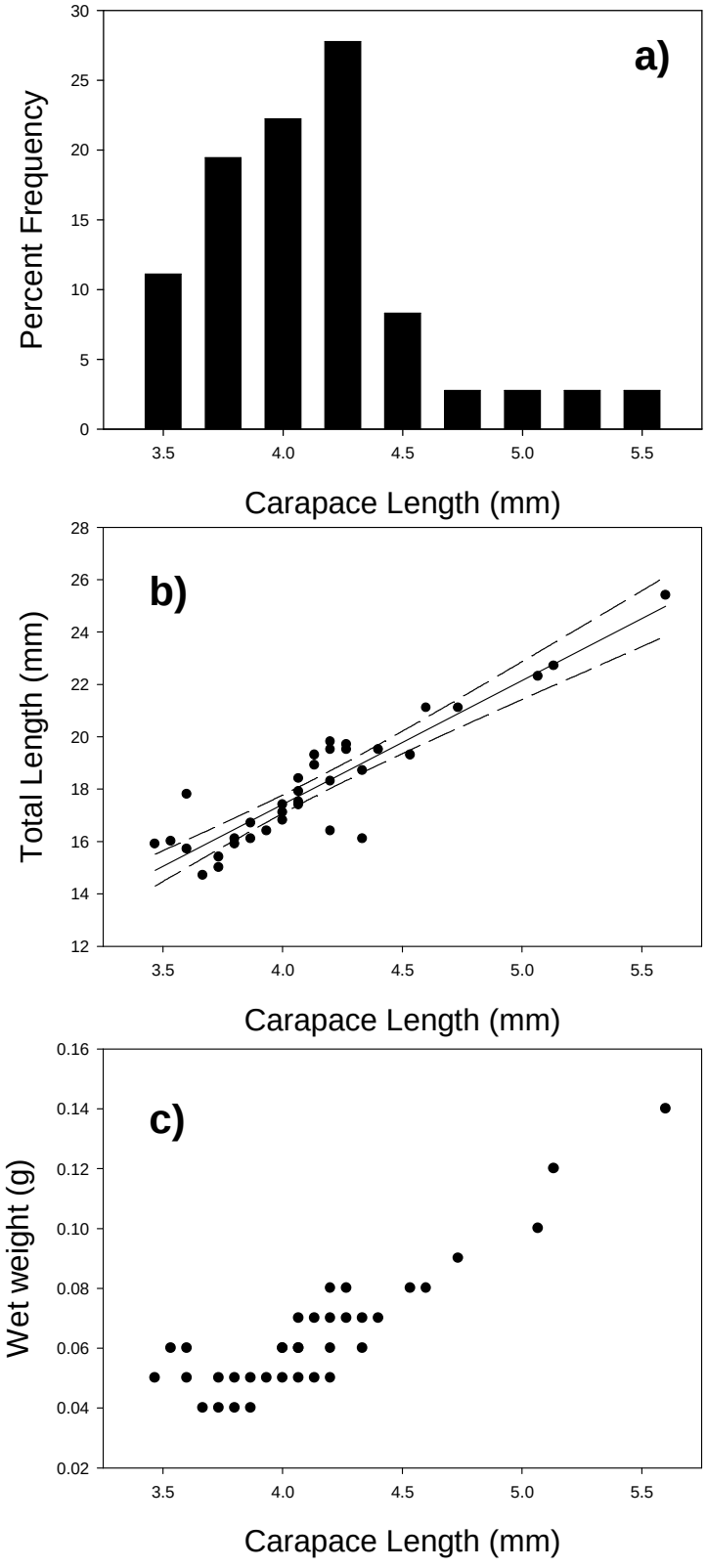

a)
Percent Frequency
30
25
20
15
10
5
0
3.5
4.0
4.5
5.0
5.5
Carapace Length (mm)
b)
Total Length (mm)
28
26
24
22
20
18
16
14
12
3.5
4.0
4.5
5.0
5.5
Carapace Length (mm)
c)
Wet weight (g)
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
3.5
4.0
4.5
5.0
5.5
Carapace Length (mm)

Figure 2.

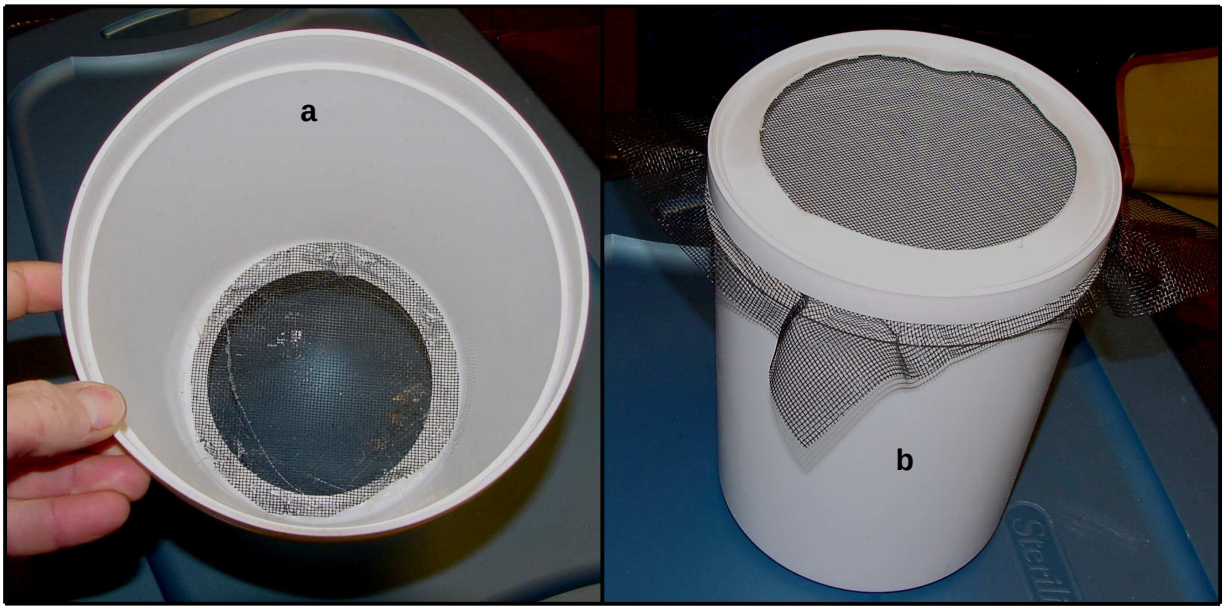


Figure 3.

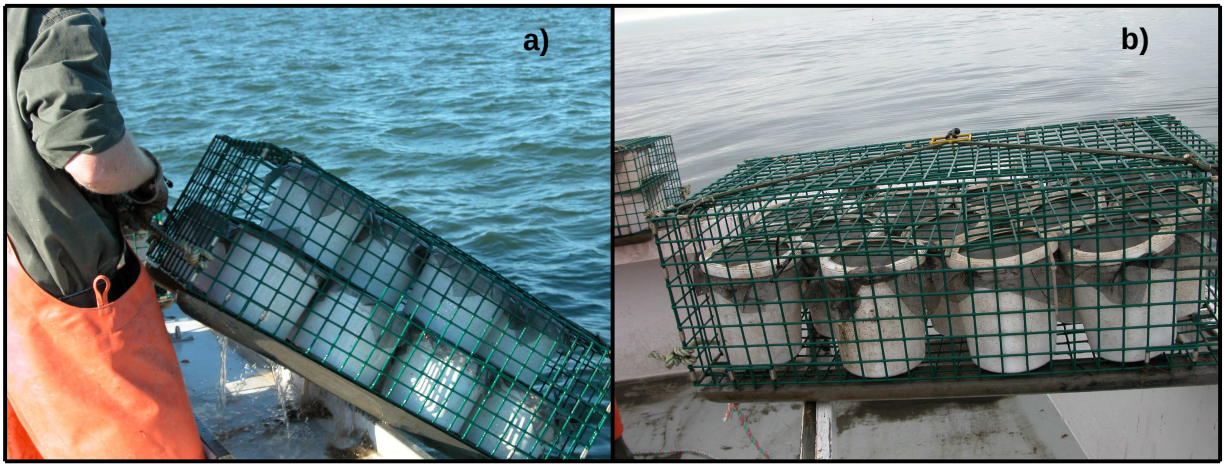


Figure 4.

Figure 5.

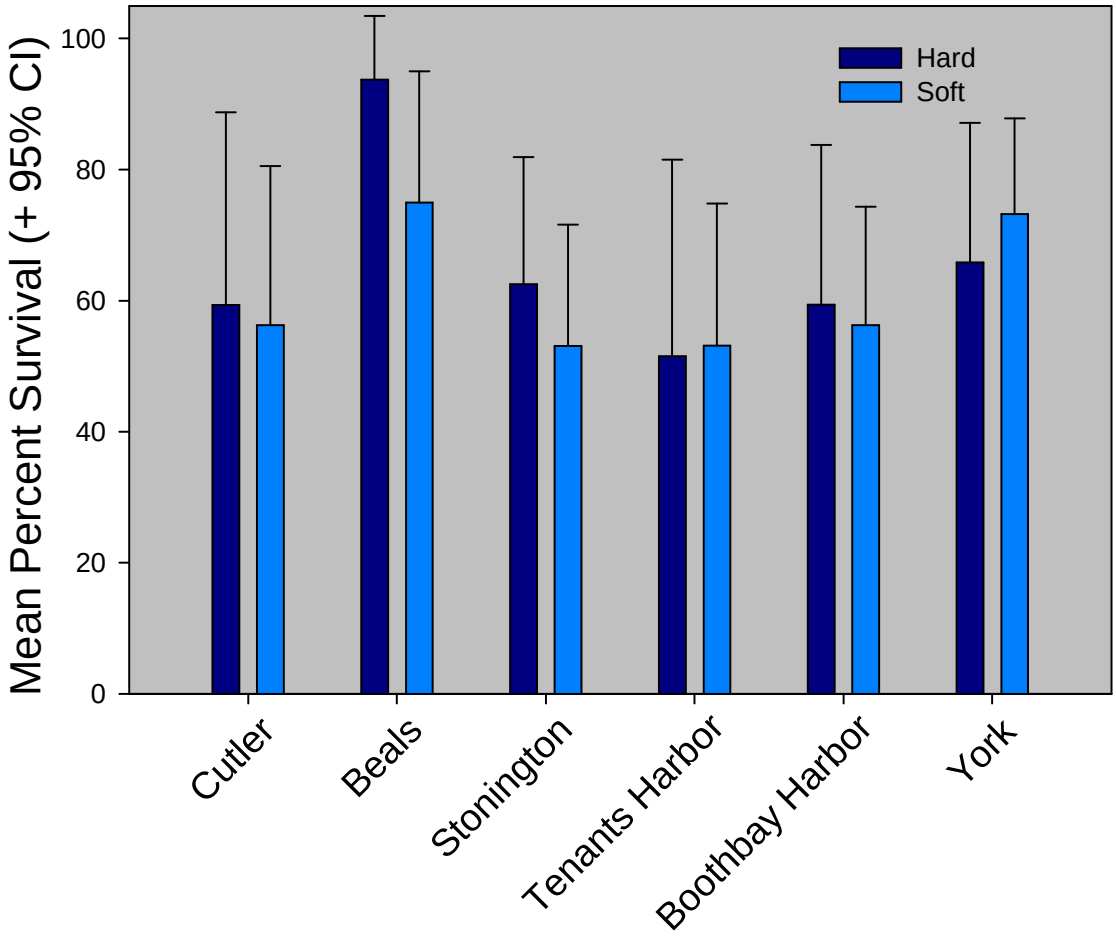

Mean Percent Survival (+ 95% CI)
100
80
60
40
20
0
Hard
Soft
Cutler
Beals
Stonington
Tenants Harbor
Boothbay Harbor
York

Figure 6.

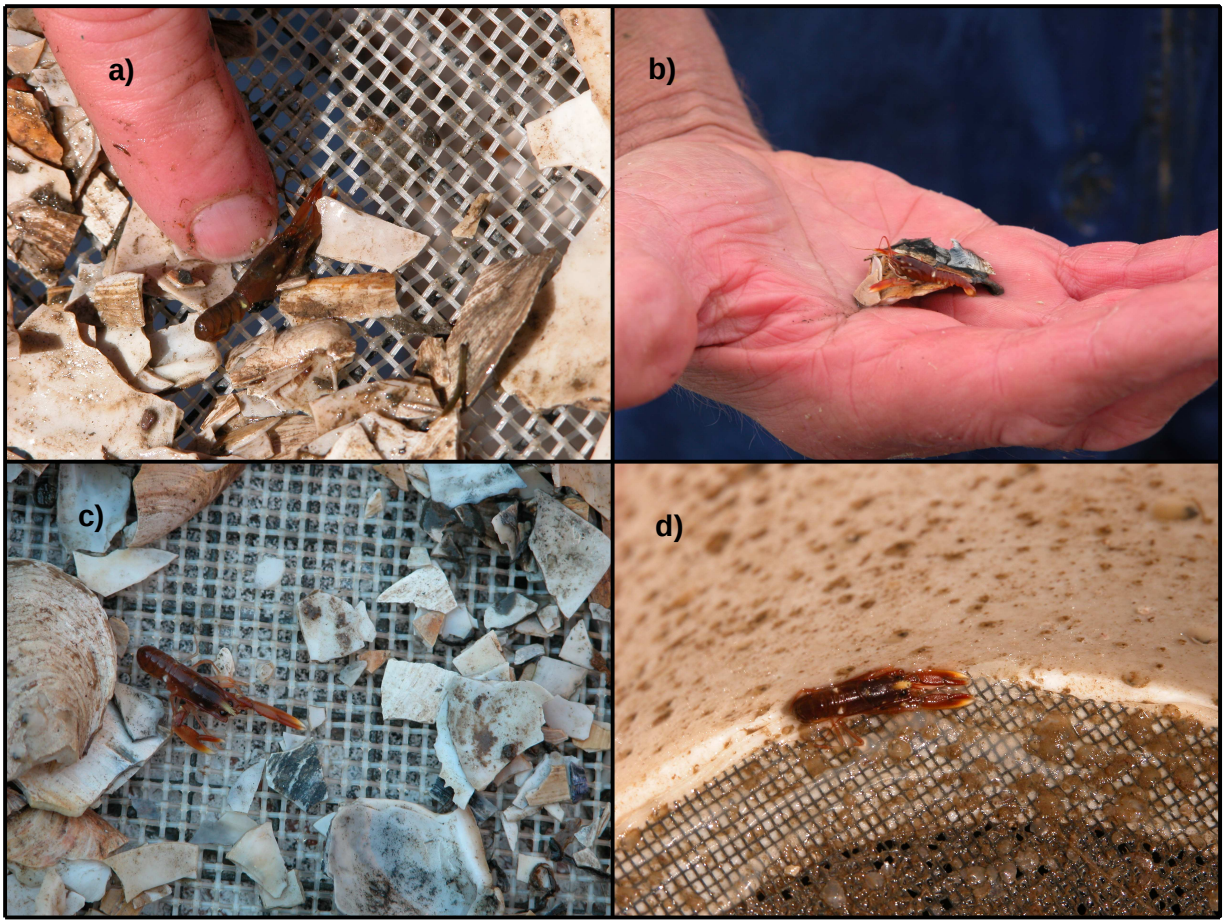


Figure 7.

Figure 8.

Figure 9.

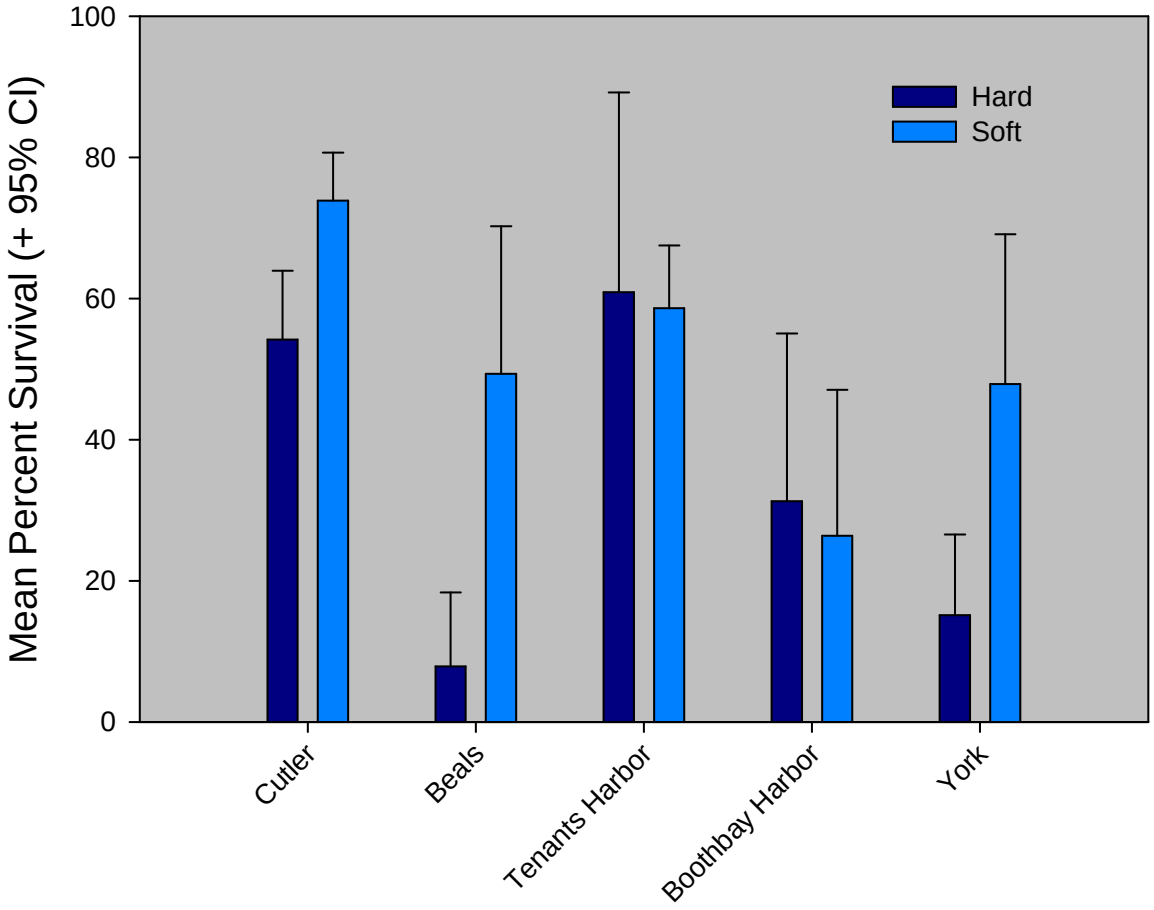

Mean Percent Survival (+ 95% CI)
100
80
60
40
20
0
Hard
Soft
Cutler
Beals
Tenants Harbor
Boothbay Harbor
York

Figure 10.

Figure 11.

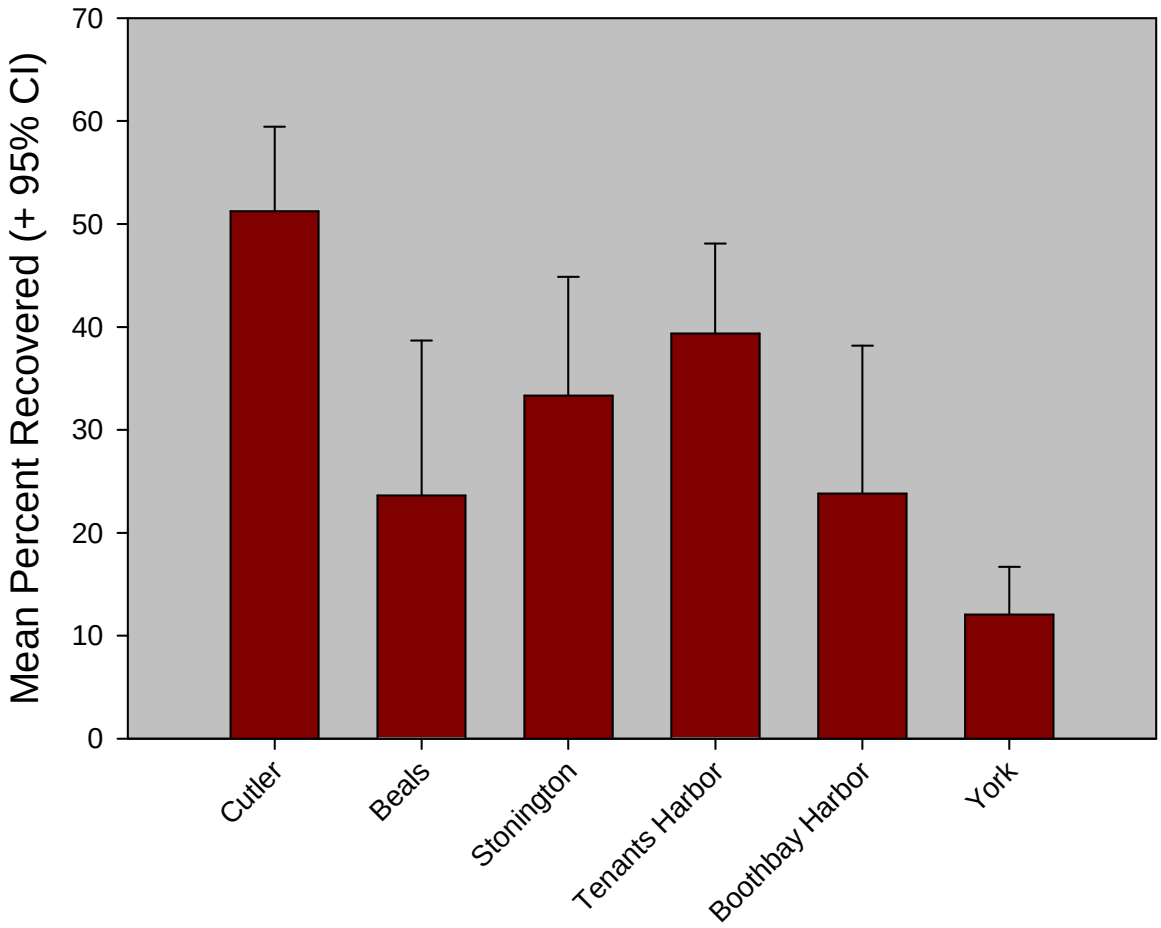

Mean Percent Recovered (+ 95% CI)
70
60
50
40
30
20
10
0
Cutler
Beals
Stonington
Tenants Harbor
Boothbay Harbor
York

Figure 12.

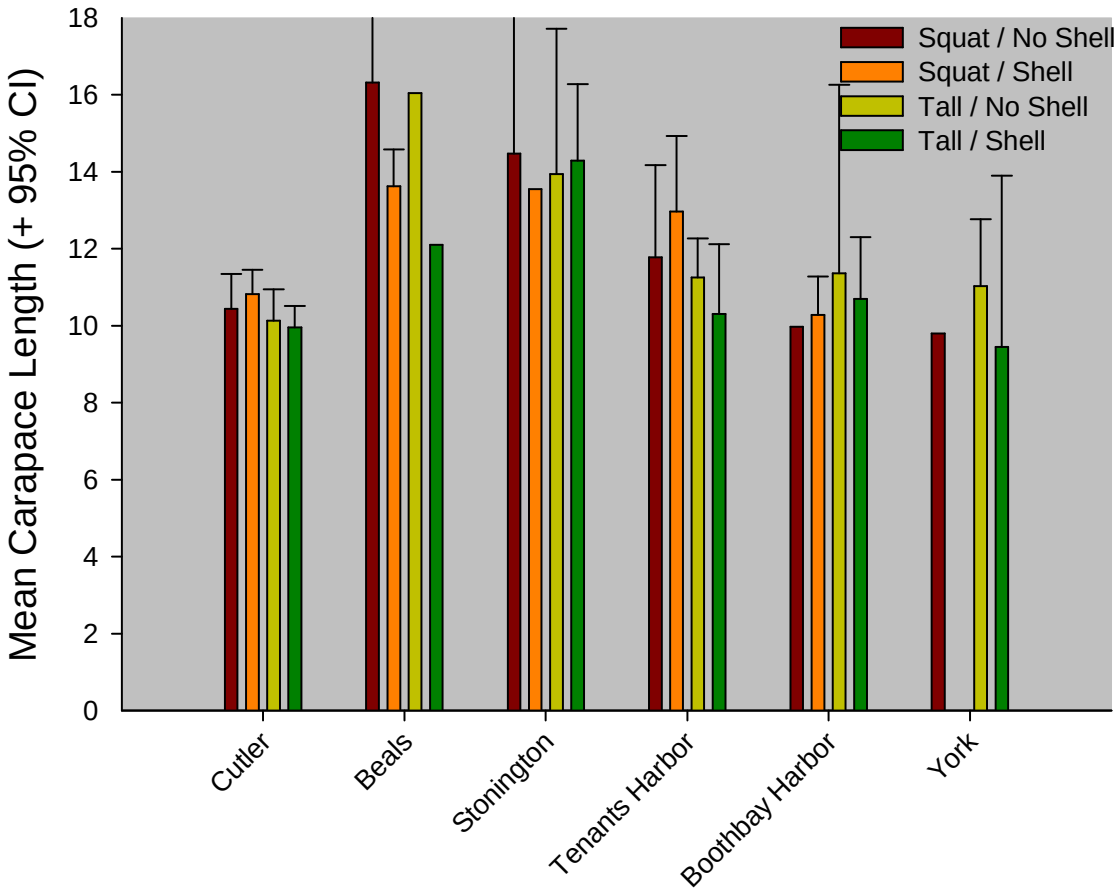

Mean Carapace Length (+ 95% CI)
18
16
14
12
10
8
6
4
2
0
Squat / No Shell
Squat / Shell
Tall / No Shell
Tall / Shell
Cutler
Beals
Stonington
Tenants Harbor
Boothbay Harbor
York

Figure 13.

Figure 14.

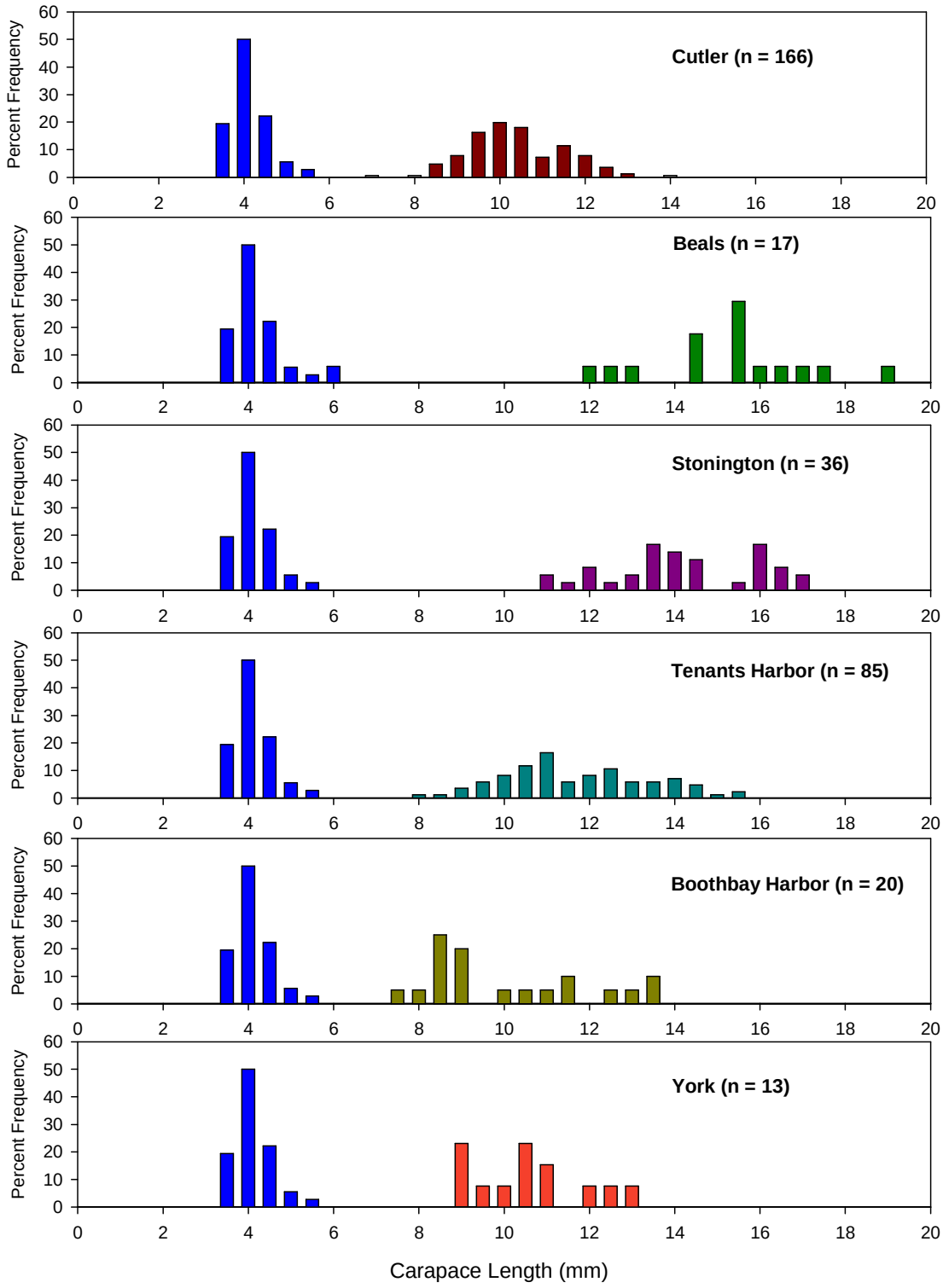

Percent Frequency
Cutler (n = 166)
Beals (n = 17)
Stonington (n = 36)
Tenants Harbor (n = 85)
Boothbay Harbor (n = 20)
York (n = 13)
Carapace Length (mm)

Figure 15.

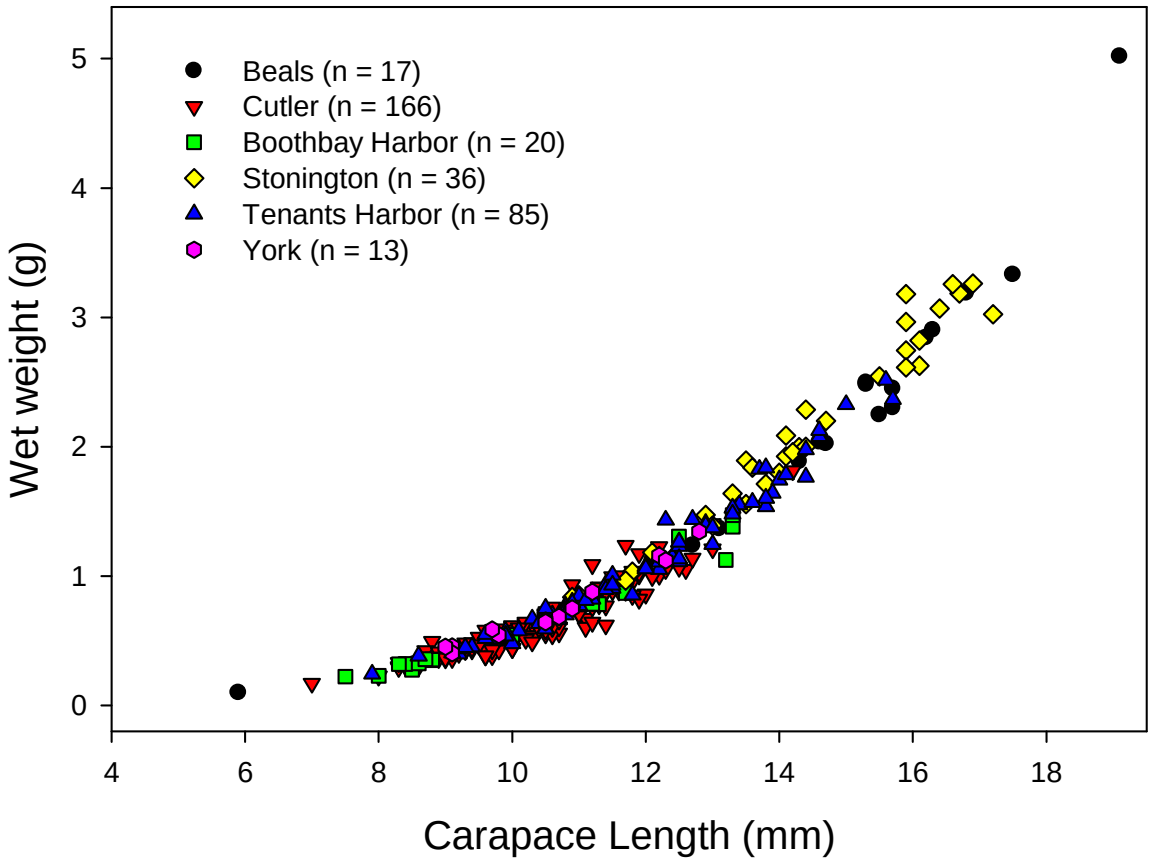

Beals (n = 17)
Cutler (n = 166)
Boothbay Harbor (n = 20)
Stonington (n = 36)
Tenants Harbor (n = 85)
York (n = 13)
Wet weight (g)
Carapace Length (mm)
0
1
2
3
4
5
4
6
8
10
12
14
16
18

Figure 16.

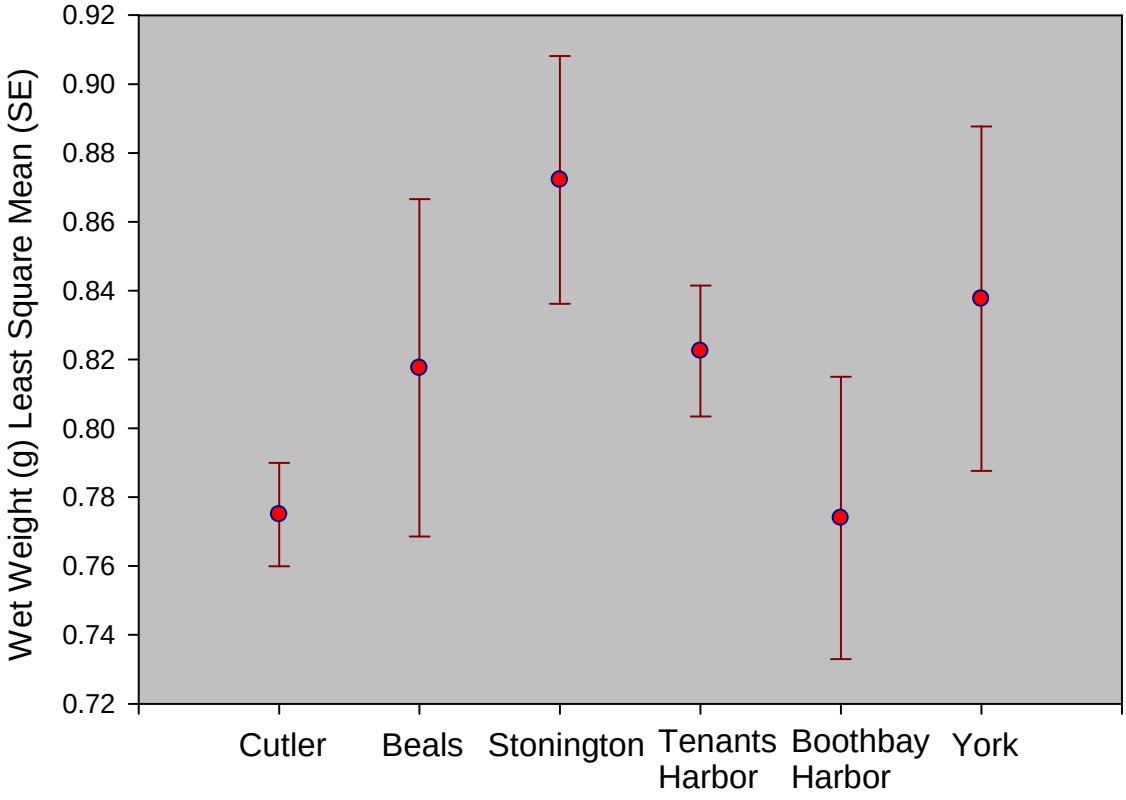

Wet Weight (g) Least Square Mean (SE)
0.92
0.90
0.88
0.86
0.84
0.82
0.80
0.78
0.76
0.74
0.72
Cutler
Beals
Stonington
Tenants Harbor
Boothbay Harbor
York

Figure 17.

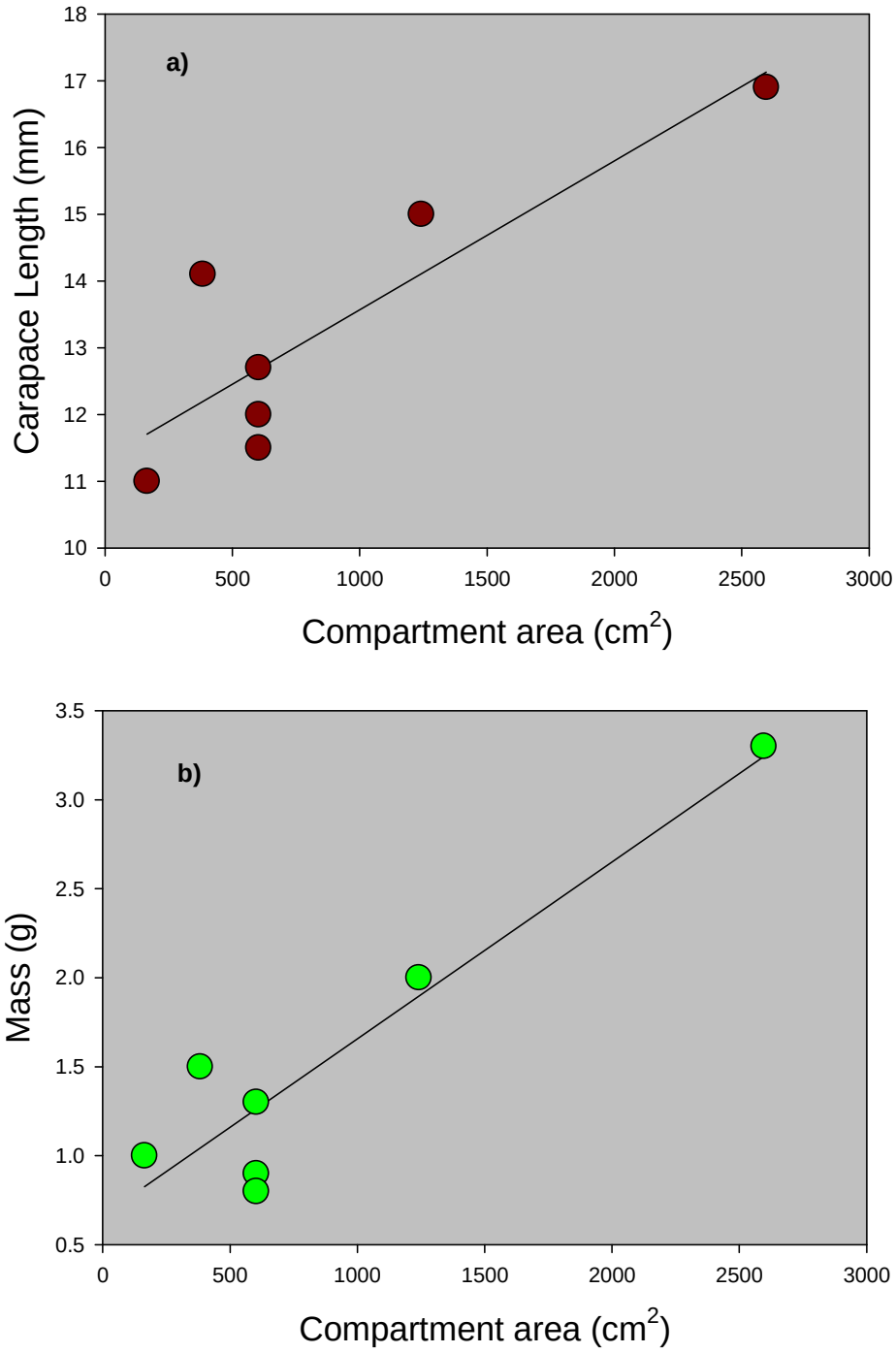
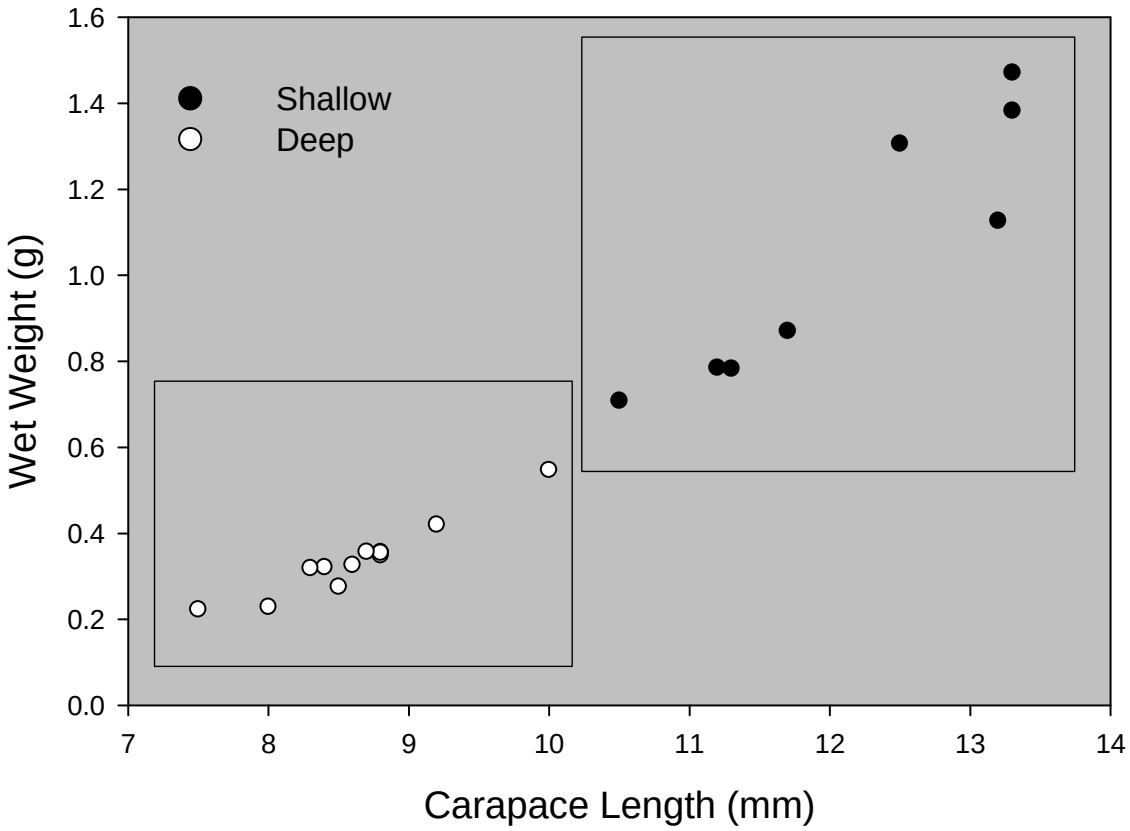

a)
Carapace Length (mm)
18
17
16
15
14
13
12
11
10
0
500
1000
1500
2000
2500
3000
Compartment area (cm2)
b)
Mass (g)
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0
500
1000
1500
2000
2500
3000
Compartment area (cm2)

Figure 18.


Shallow
Deep
Wet Weight (g)
Carapace Length (mm)
0.0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
1.6
7
8
9
10
11
12
13
14